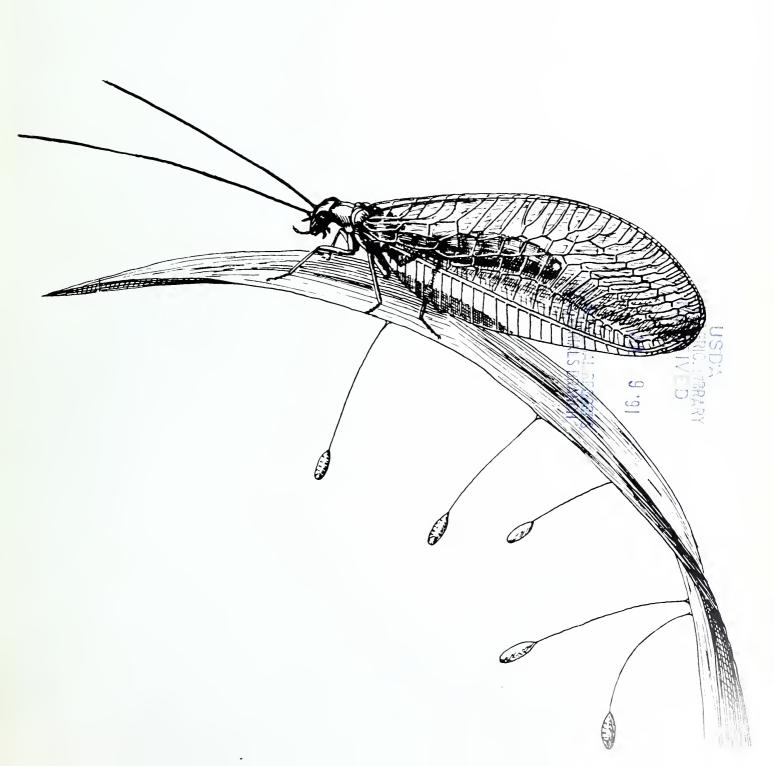


Agricultural Research Service

Miscellaneous Publication Number 1443 Insects and Mites:
Techniques for Collection
and Preservation

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Abstract

Steyskal, George C., William L. Murphy, and Edna M. Hoover, editors. 1986. Insects and mites: Techniques for collection and preservation. U.S. Department of Agriculture, Miscellaneous Publication No.1443,103 pp.

This publication describes the most effective methods and equipment for collecting, identifying, rearing, examining, and preserving terrestrial and aquatic insects and mites and for storing and caring for specimens in an insect collection. Instructions are provided for the construction of many kinds of collecting equipment, traps, rearing cages, and storage units. Keys are included for the identification of the classes of Arthropoda and the orders of insects. Each order of insects is treated separately, and techniques unique to each order are discussed. Instructions are also given for obtaining identifications of insects and mites from the U.S. Department of Agriculture and on how to pack specimens for shipping. An extensive list of references is included.

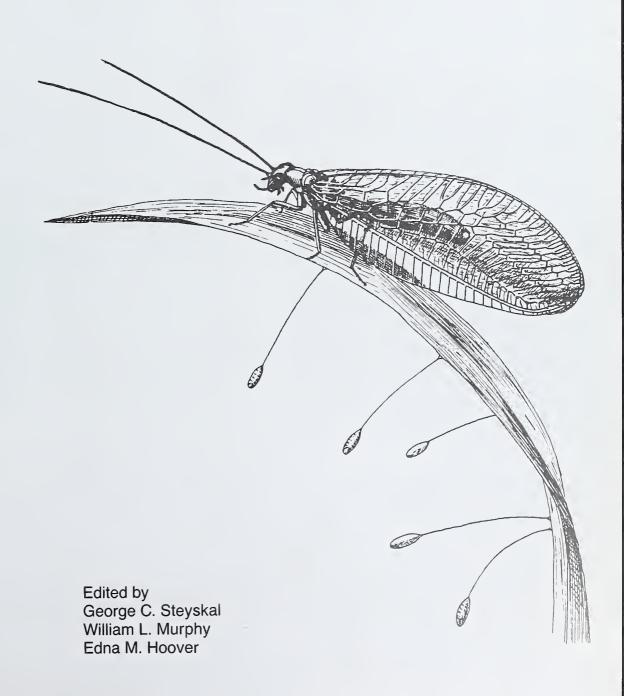
KEYWORDS: Acari, Apterygota, Arachnida, Arthropoda, attractants, baits, Chilopoda, Crustacea, Diplopoda, insect classification, insect collecting and collections, insect collecting equipment, insect collecting techniques, insect identification, insect taxonomy, Insecta, mite classification, mite collecting and collections, mite collecting equipment, mite collecting techniques, mite identification, mite taxonomy, mounting, Pauropoda, pheromones, preservation of insects, preservation of mites, Pterygota, rearing, rearing conditions, Symphyla, traps.

United States Department of Agriculture

Agricultural Research Service

Miscellaneous Publication Number 1443

Insects and Mites: Techniques for Collection and Preservation



Foreword

This publication is based on "Collection and Preservation of Insects," U.S. Department of Agriculture Miscellaneous Publication 601, by P. W. Oman and Arthur D. Cushman (both retired) of the former Insect Identification and Parasite Introduction Research Branch, Agricultural Research Service. It was issued in 1948, reprinted several times, and slightly revised in 1967. It was intended to serve the needs of students, professional entomologists, amateur collectors, and others interested in the collection and study of insects. The present publication is intended to serve similar needs, but it has been expanded greatly to include important new techniques and equipment developed since the 1950's. Changes in almost all aspects of the study and application of entomology have made the collection and preservation of insects far more complex than in 1948. Concomitantly, since the literature on these subjects has become extensive, a list of selected references has been added.

The scope of this publication has also been broadened to include mites because of the increasing awareness of their economic importance to agriculture and because they are an integral part of the systematics research and service program of the Biosystematics and Beneficial Insects Institute (BBII), USDA.

This publication is also intended to serve as a guide for the proper preparation of insects and mites that may be submitted to the BBII for identification. Keys have been added for the identification of the major orders of insects and mites to aid in determining what has been collected and how best to preserve it, because preservation techniques vary for different kinds of insects and mites. The instructions in this publication are necessarily brief; however, they should provide sufficient guidance to allow the reader to make and maintain a collection of insects and mites in a condition suitable for study. The collector requiring additional or more specialized information may wish to refer to the list of selected publications, many of which may be found in public or university libraries. Specific questions or comments on the USDA insect and mite identification service may be addressed to the Director, Biosystematics and Beneficial Insects Institute, Beltsville Agricultural Research Center, Beltsville, Md. 20705.

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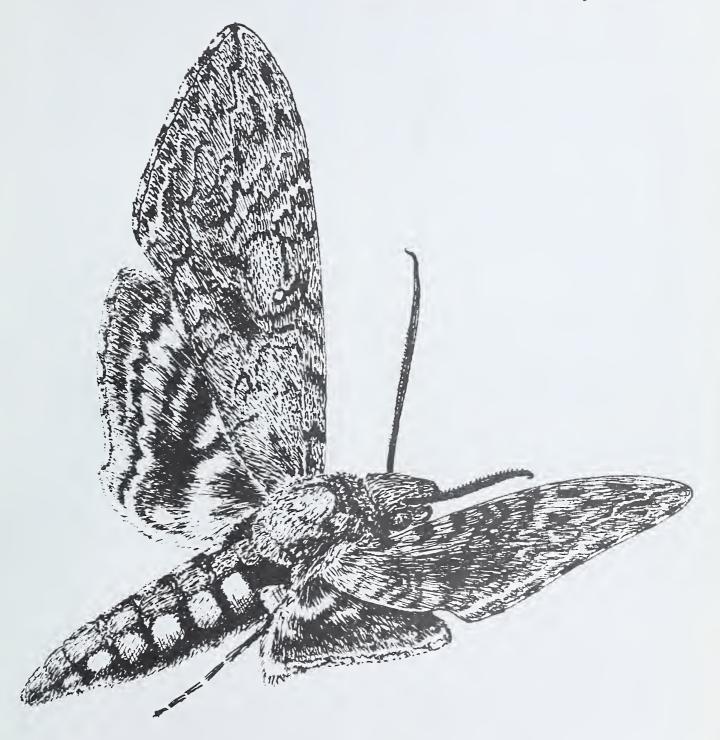
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Basic Tools and General Techniques



Insects and Mites: Techniques for Collection and Preservation

Edited by George C. Steyskal, William L. Murphy, and Edna M. Hoover¹

Part 1 Basic Tools and General Techniques

Reasons for Collecting Insects and Mites

The number and variety of insects and mites known today stagger the imagination. Not only are there more kinds or species of insects than of all other animals combined, but insects and mites can be found in almost every conceivable environment. Their habits vary from parasitic to free-living and from beneficial to highly destructive. The destructive forms can cause enormous losses in terms of diseases and damage to food, clothing, and other materials of value to humans and can injure people or transmit disease to them directly.

Because of the potential destructiveness of some species of insects and mites, it is essential that outbreaks of both old and new pests be detected and that population sizes be estimated as rapidly as possible. Correct identification of a newly detected pest is of the utmost importance, because the scientific name is the key to all known information about the insect, its habits, its behavior, and its potential threat or benefit to human welfare.

Insects and mites can and should be observed in their natural environments, but most of them, especially the many small ones, must be collected and properly preserved before they can be identified. Because correct identification seldom is easy, it is important that specimens be preserved in the best condition possible. The identification of a particular insect or mite usually requires examination of minute details of its anatomy with the aid of a hand lens or microscope. If these details on a specimen are concealed or missing because of improper handling or preservation, identification is impossible, and information about the species to which it belongs cannot be made available. Therefore, adequate preservation and proper labeling of specimens are essential to their identification.

The methods used and when and where to collect depend on the purpose of the collecting. Insects and mites may be collected to obtain general study mate-

¹G. C. Steyskal (retired), Biosystematics and Beneficial Insects Institute (BBII), c/o National Museum of Natural History, Smithsonian Institution, Washington, D.C. 20560; W. L. Murphy and E. M. Hoover (retired), BBII, Beltsville Agricultural Research Center, Beltsville, Md. 20705. rial, as for a school course or for personal interests (hobby); to survey for the presence or abundance (relative or absolute numbers) of pests; to acquire material for biological, physiological, ecological, and other types of studies; or to obtain specimens for other purposes that may demand various special collecting methods.

What to Collect

What to collect depends on the purpose for which the material is intended. When important pest insects and mites need to be identified, they should be collected in large numbers if at all feasible. A sample of 20 specimens should be considered the minimum, and even larger numbers are desirable. If adults and immatures are present, specimens should be collected of all life stages. Excess specimens can be discarded or exchanged, but it is not always possible to collect additional specimens when needed. Frequently insects and mites cannot be identified accurately from immature stages, and it is then necessary to rear them to the adult stage to obtain a precise identification. Photographers should collect the specimens they photograph if positive identification is desired; minute, critical diagnostic characters often are not depicted in photographs. If specimens are destined for display cases, it may be important to collect a sample of the host plant for the display. When collecting gall-producing insects and mites, it is essential to obtain the gall as well.

Many persons starting a collection attempt to collect every specimen they find. Biology students in high school and college are often required to collect specimens from as many orders or groups as possible. The experience and knowledge gained in making a general collection are of value in helping the collector decide on a specialty. However, with so many different kinds of insects from which to choose—over 80,000 described species in North America alone—most persons find that concentrating eventually on 1 or 2 of the major insect or mite groups is desirable. Specimens other than those in a chosen group may still be collected for exchange with other collectors.

References: 265, 380.2

Equipment and Collecting Methods

Collecting methods may be divided into two broad categories. In the first the collector actively searches out the insects, using nets, aspirators, beating sheets, or whatever apparatus suits his or her particular needs. In the second, the collector participates passively and permits traps to do the work. Both approaches may be used simultaneously, and both are discussed in the following pages. To obtain as many specimens as possible, use as many different collecting methods as possible, especially when briefly visiting an interesting area.

²See list of Selected References, page 80.

The simplest method of collecting is to pick up specimens by hand. For many reasons, however, such as difficulties in capturing insects and injury or discomfort that the insects may cause the collector, various kinds of equipment and special methods are needed. Those described here have general application; it is expected that the collector will make some adaptations to fit his or her own purposes and resources. For additional information, especially concerning the use of specialized techniques, consult the list of references.

References (general): 9, 13, 15, 16, 43, 46, 53, 69, 70, 80, 113, 132, 238, 240-242, 261, 266, 284, 285, 330, 334, 335, 345, 381, 394, 399, 431-433, 436.

The equipment used to assemble an insect or mite collection need not be elaborate or expensive. In many instances, a collecting net (see p. 5) and several killing bottles (see p. 7) will suffice; however, additional items will permit more effective sampling of a particular fauna. Many collectors carry a bag or wear a vest in which they store equipment. The following items usually are included in the general collector's bag:

- (1) Forceps. Fine, lightweight forceps are recommended; if sharp-pointed forceps are used, care must be taken not to puncture specimens. If possible, grasp specimens with the part of the forceps slightly behind the points.
- (2) Vials containing alcohol or other preservatives (see p. 21).
- (3) Killing bottles of various sizes.
- (4) Small boxes for storing specimens after their removal from killing bottles. These may be made of cardboard, plastic, or metal and should be partly filled with soft tissue to keep specimens from rolling about. Do not use cotton because specimens become entangled in the fibers and may become virtually impossible to extricate without damage.
- (5) Small envelopes for temporary storage of delicate specimens.
- (6) One or more aspirators (see p. 8).
- (7) Absorbent tissue for use in killing bottles and aspirators.
- (8) Notebook and writing equipment for jotting down notes and label data.
- (9) A strong knife for opening galls, seed pods, twigs, etc.
- (10) A small, fine brush (camel's hair is best) for picking up minute specimens. Moisten the tip; tiny specimens will adhere to it and may be transferred to a killing bottle or vial.

- (11) Bags for storing plant material, rearing material, or Berlese samples (see p. 10). For collecting much plant material, a botanist's vasculum or tin box is advisable.
- (12) A hand lens; one worn on a lanyard is convenient.

This list may be modified according to the special kinds of insects or mites to be collected. A small digging tool or trowel may be useful for collecting insects from soil or for gathering Berlese samples and a heavy knife or small hatchet for searching under bark or in decaying logs. A plant press should be availabe to prepare plant specimens for determination or as voucher specimens, especially when leaf-mining insects are being studied. When collecting at night, have a flashlight or headlamp; the latter is especially useful because it leaves the hands free.

Much of the equipment may be obtained from ordinary sources, but equipment especially designed for insect collecting often must be bought from special supply houses. Their addresses may be found in the yellow pages of telephone directories under "Biological Laboratory Supplies" or "Laboratory Equipment and Supplies." Inquiry to biology departments of high schools and universities may also be helpful, and biological and entomological journals often carry advertisements of equipment suppliers. Inasmuch as these firms are located in many parts of the country and change names and addresses fairly often, it is not practical to list them here. The faculty members of the local university's biology department are usually willing and in the best position to recommend a supplier in their area.

Collecting Nets

Collecting nets come in three basic forms: Aerial, sweeping, and aquatic. The first is designed especially for collecting butterflies and other flying insects. Both the bag and handle are relatively lightweight. The sweeping net is similar to the aerial net but is stronger and has a more durable bag to withstand being dragged through dense vegetation. Aquatic nets are used for gathering insects from water and are usually made of metal screening or heavy scrim with a canvas band affixed to a metal rim. A metal handle is advisable because wooden ones may develop slivers after repeated wetting. The net you choose depends on the kind of insects or mites you wish to collect.

Several kinds of nets, including collapsible models with interchangeable bags, are available from biological supply houses, but anyone with a little mechanical ability can make a useful net. The advantage of a homemade net is that the size and shape can be adapted to the needs of the user, to the kind of collecting intended, and to the material available, which need not be expensive. These materials include—

(1) Piece of heavy (8-gage) steel wire for the rim, bent to form a ring 30-38 cm in diameter (fig. 1, A). Small nets 15 cm or so in diameter sometimes are useful, but

nets larger than 38 cm are too cumbersome for most collecting.

- (2) Dacron or other strong, light fabric through which air can flow freely. Brussels netting is best but may be difficult to obtain; otherwise nylon netting, marquisette, or good quality cheesecloth can be used, but the last snags easily and is not durable. The material should be folded double and should be 1.5-1.75 times the rim diameter in length (fig. 1, *B*). The edges should be double-stitched (French seams).
- (3) Strip of muslin, light canvas, or other tightly woven cloth long enough to encircle the rim. The open top of the net bag is sewn between the folded edges of this band to form a tube through which the wire rim is inserted (fig. 1, C).
- (4) Straight hardwood dowel about 19 mm in diameter and 105-140 cm long (to suit the collector). For attachment of the rim to the handle, a pair of holes of the same diameter as the wire are drilled opposite each other to receive the bent tips of the wire, and a pair of grooves as deep and as wide as the wire are cut from each hole to the end of the dowel to receive the straight part of the wire (fig. 1, *D*).
- (5) Tape or wire to lash the ends of the rims tightly into the grooves in the end of the handle. This may be electrician's plastic tape or fiber strapping tape commonly used for packaging. If wire is used, the ends should be bound with tape to secure them and to keep them from snagging. A close-fitting metal sleeve (ferrule) may be slipped over the rim ends and held in place with a small roundheaded screw instead of tape or wire lashing.

After the net has been placed on the rim, the ends of the band should be sewn together and the rim ends fastened to the handle. The other end of the handle should be filed to remove sharp edges. The net is then ready for use (fig. 1, *E*).

Efficient use of a net is gained only with experience. Collection of specimens in flight calls for the basic stroke—swing the net rapidly to capture the specimen, then follow through to force the insect into the very bottom of the bag. Twist the wrist as you follow through so the bottom of the bag hangs over the rim (fig. 2); this will entrap the specimen. If the insect alights on the ground or other surface, it may be easier to use a downward stroke, quickly swinging down on top of the insect. With the rim of the net in contact with the ground to prevent the specimen from escaping, hold the tip of the bag up with one hand. Most insects will fly or crawl upward into the tip of the bag, which can then be flipped over the rim to entrap the specimen.

Sweeping the net through vegetation, along the sand and seaweed on beaches, or up and down tree trunks will catch many kinds of insects and mites. The aerial net may be used in this way, but the more durable sweeping net is recommended for such rough usage. After sweeping with the net, a strong swing will bring

anything in the bag to the bottom, and then by immediately grasping the middle of the net with the free hand, the catch will be confined to a small part of the bag. Only the most rugged sweeping net may be used through thistles or brambles. Even some kinds of grasses, such as sawgrass, can quickly ruin a net. Burs and sticky seeds are also a serious problem.

The catch may be transferred from the bag to a killing jar in one of several ways. Single specimens are transferred most easily by lightly holding them in a fold of the net with one hand while inserting the open killing jar into the net with the other. While the jar is still in the net, cover the opening until the specimen is stupefied; otherwise, it may escape before the jar can be removed from the net and closed. To prevent a butterfly from damaging its wings by fluttering in the net, squeeze the thorax gently through the netting when the butterfly's wings are closed. Experience will teach you how much pressure to exert; obviously, pinching small specimens of any kind is not recommended. When numerous specimens are in the net after prolonged sweeping, it may be desirable to put the entire tip of the bag into a large killing jar for a few minutes to stun the insects. They may then be removed and desired specimens placed separately into a killing jar, or the entire mass may be dumped into a killing jar for later sorting. These methods of mass collecting are especially adapted to obtaining small insects not readily recognizable until the catch is sorted under a microscope.

Removal of stinging insects from a net may be a problem. They will often crawl toward the rim of the bag and may be made to enter a killing jar held at the point where they crawl over the rim. However, many insects will fly as soon as they reach the rim, and a desired specimen may be lost. A useful method is to trap the insect in a fold of the net, carefully keeping a sufficient amount of netting between fingers and insect to avoid being stung. This fold of the net can then be inserted into the killing jar to stun the insect. After a few moments, it should be safe to remove the insect from the net and transfer it to a killing jar. If the stunned insect clings to the net and does not fall readily into the jar, use forceps or pry the insect loose with the jar lid or a small stick—not with your fingers.

Aerial nets made of dacron or nylon may be used to sweep insects from water if an aquatic net is not at hand. The netting will dry quickly if swept strongly through the air a few times; however, it should not be used again until thoroughly dry, or other specimens, especially butterflies, may be ruined. A number of special modifications are necessary to adapt a net for aquatic collecting.

For specialized collecting, nets can be attached to the ends of beams that are rotated about their midlength by a motor drive. Nets also can be adapted to be towed by or mounted on vehicles.

References: 108, 226, 366, 371, 413, 455; vehicle-mounted net: 6, 19, 170, 188, 196, 208, 248, 254, 284, 371, 425, 426.

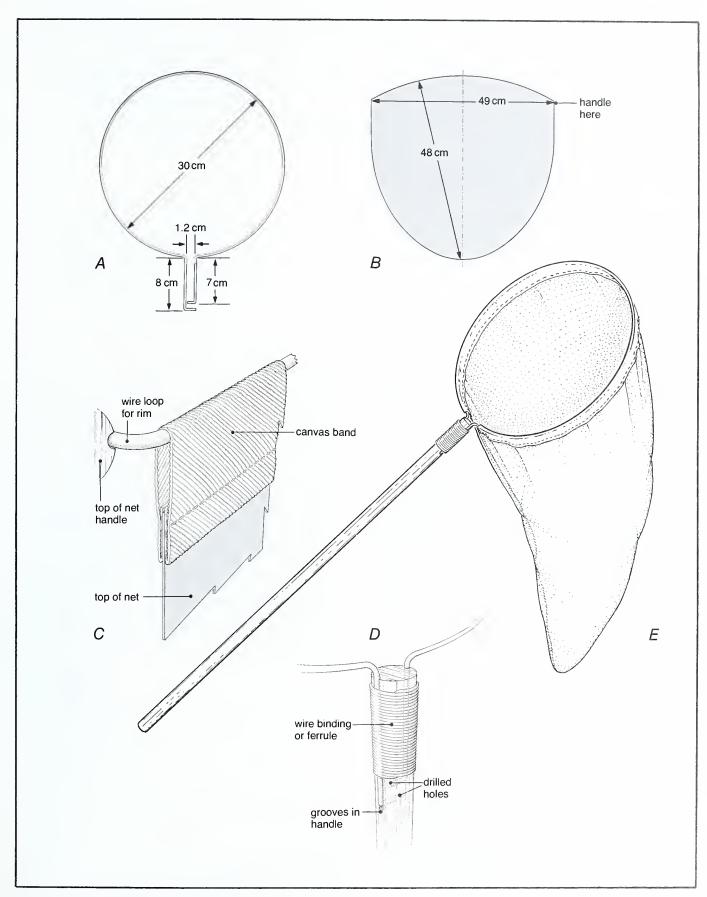
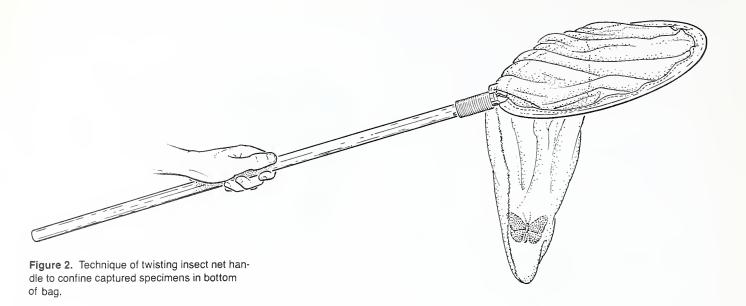


Figure 1. Construction of a sweeping net: *A*, Steel wire loop; *B*, pattern for cutting net bag; *C*, top of net sewed to canvas band that is fitted

over wire loop; *D*, end of net handle showing grooves and holes into which arms of wire loop fit; *E*, completed net with wooden handle.



Killing Jars or Bottles

Any heavy, wide-mouthed glass jar or bottle with a tight-fitting stopper or metal screw top may be used as a killing container. Olives frequently are sold in bottles that make convenient killing containers. Tops that may be removed with only a quarter turn often are preferred but may not be obtained readily. The killing agent used may be any of various liquids or cyanide granules. Liguid killing agents generally are considered less effective but safer to use than cyanide, but some of them are known to accumulate in human tissue after repeated or prolonged exposure. Despite its extreme toxicity, cyanide is a noncumulative poison, and brief exposure to the fumes, as inevitably occurs when opening jars to insert or remove specimens, does no permanent harm so far as is known. Never deliberately inhale the fumes, even momentarily. All killing agents are to some extent hazardous to human health. All killing jars or bottles should be clearly labeled "POISON" and should be kept away from persons unaware of their danger.

Liquid Killing Agents. Jars for use with liquid killing agents are prepared in one of two ways. One way (fig. 3, A) is to pour about 2.5 cm of plaster of paris mixed with water into the bottom of the jar and allow the plaster to dry without replacing the lid. Enough of the killing agent is then added to saturate the plaster; any excess should be poured off. This kind of jar can be recharged merely by adding more killing agent. The second method is to place a wad of cotton or other absorbent material in the bottom of a jar, pour enough liquid killing agent into the jar to nearly saturate the absorbent material, and then press a piece of stiff paper on it or a cardboard cut to fit the inside of the jar tightly. The paper or cardboard acts as a barrier between the insect and the killing agent, keeping the latter from evaporating too rapidly and also preventing the specimen from becoming entangled in loose fibers.

Among the liquid killing agents are ethyl acetate $(CH_3CO_2 \cdot C_2H_5)$, carbon tetrachloride (CCI_4) , ether

(diethyl ether, C_2H_5 · O · C_2H_5), chloroform (CHCl₃), and ammonia water (NH₄OH solution).

Ethyl acetate is recommended by many as the most satisfactory liquid killing agent. Its fumes are less toxic to humans than those of the other substances. Although it usually stuns insects quickly, it kills them slowly. Specimens that appear dead may revive if removed from the killing jars too soon, but a compensating advantage is that most specimens may be left in an ethyl acetate killing jar for several days and still be limp. If the ethyl acetate is allowed to evaporate from the specimens, they will harden. A killing jar with ethyl acetate is preferred by many entomologists over the cyanide jar (see p. 7), especially for infrequent use.

Carbon tetrachloride was once very popular as a liquid killing agent because of its ready availability as a spot remover for clothes and its nonflammability. It is no longer recommended, however, because it is a carcinogen and cumulative liver toxin, and specimens killed with it become brittle and difficult to pin.

Ether and chloroform are both extremely volatile and flammable and should not be used near an open flame or lighted cigarette. Their high volatility makes them serviceable in a killing jar for only a short time. Perhaps the greatest hazard with chloroform is that even when stored in a dark-colored jar, it eventually forms the extremely toxic gas phosgene (carbonyl chloride, COCl₂). Chloroform, however, is useful when other substances cannot be obtained. It stuns and kills quickly but has the disadvantage of stiffening specimens.

Ammonia is irritating to humans, does not kill very effectively, and spoils the colors of many specimens. It is, however, readily available and will serve in an emergency. Ammonium carbonate, a solid but volatile substance, is also useful.

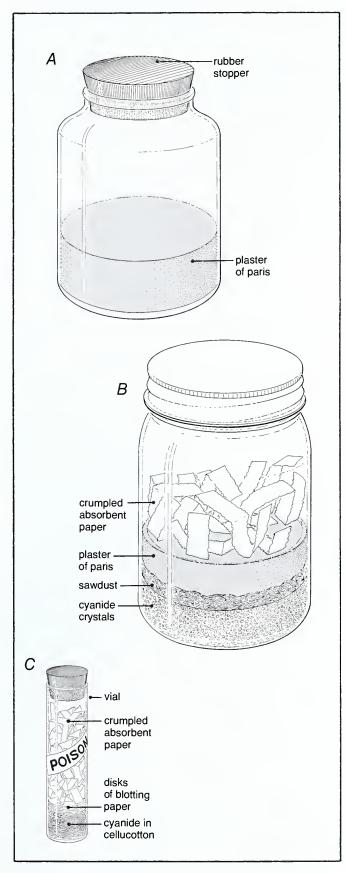


Figure 3. Widemouthed killing jars for use with liquid (A) and solid (B) killing agents. C, Vial-type cyanide bottle for small insects with all-important "POISON" label.

Solid Killing Agents. The solid killing agents used in killing jars are the cyanides—potassium cyanide (KCN), sodium cyanide (NaCN), or calcium cyanide [Ca(CN)₂]. Handle all cyanides with extreme care. They are dangerous, rapid-acting poisons with no known antidote. If even a single grain touches the skin, wash immediately with water. To avoid handling the cyanide and having to find a safe place to store or dispose of surplus crystals, you may be able to find a chemist, pharmacist, or professional entomologist to make the killing jar for you. If this is not feasible, use utmost care in following the instructions given here.

To make a cyanide killing jar or bottle, place about 15 mm of cyanide crystals in the bottom (fig. 3, B). Potassium cyanide is best; sodium cyanide is as effective but is hygroscopic, that is, it absorbs water and makes the jar wet; and calcium cyanide is seldom available. Cover the crystals with about 10 mm of sawdust and add about 7 mm of plaster of paris mixed with water to form a thick paste, working quickly before the plaster solidifies. Then add crumpled absorbent paper to prevent water condensation on the inside glass surface. Instead of the plaster of paris, a plug of paper or cardboard may be pressed on top of the sawdust. Be sure that it fits tightly. When ready to use after a few hours, place several drops of water on the plaster or paper plug. In an hour or so, enough fumes of hydrocyanic acid will have been produced to make the jar operative. Do not test this by sniffing the open jar.

Another substance that has been recommended as a killing agent is dichlorvos (2,2-dichloroethenyl dimethyl phosphate), also called DDVP, Vapona, Nogos, Herkol, and Nuvan. Polyvinyl chloride (PVC) resin, impregnated with this chemical and sold commercially as No-Pest Strips, is long lasting and somewhat less dangerous to use than the other killing agents, but its time-release aspect allows only small quantities of the active agent to be released, quantities too small to kill quickly. PVC-impregnated dichlorvos is therefore occasionally useful only as a killing agent in traps.

Every killing jar or bottle should be clearly and prominently labeled "POISON" (fig. 3, C). The bottom must be covered with tape, preferably cloth, plastic, or clinical adhesive tape, to cushion the glass against breakage and to keep its dangerous contents from being scattered if the container breaks.

Killing jars or bottles will last longer and give better results if the following simple rules are observed:

- (1) Place a few narrow strips of absorbent paper in each jar or bottle to keep it dry and to prevent specimens from mutilating or soiling each other. Replace the strips when they become moist or dirty. This method is useful for most insects except Lepidoptera, which are too difficult to disentangle without damage.
- (2) Do not leave killing jars in direct sunlight as they will sweat and rapidly lose their killing power.

- (3) If moisture condenses in a jar, wipe it dry with absorbent tissue.
- (4) Keep delicate specimens in separate jars so that larger specimens will not damage them.
- (5) Do not allow a large number of specimens to accumulate in a jar unless it is to be used specifically for temporary storage.
- (6) Do not leave insects in cyanide jars for more than a few hours. The fumes will change the colors of some insects, especially yellows to red, and specimens will generally become brittle and difficult to handle.
- (7) If it is necessary to keep insects in killing jars for more than several hours, place the specimens in another container and store them in a refrigerator.
- (8) Keep butterflies and moths in jars by themselves so that their hairs and scales will not ruin other kinds of insects
- (9) Never test a killing jar by smelling its contents.
- (10) Old jars that no longer kill quickly should be recharged or disposed of by burning or burying. A cyanide jar that has become dry may be reactivated by adding a few drops of water.

Spray-dispensed insecticides may be used, if not to kill specimens, to at least 'knock them down' into a container from which they may be picked up. If they are directed into a container topped with a funnel, they may be allowed to revive and treated further as desired (see ref. 82).

References: 16, 82, 138, 268, 342, 352, 447.

Aspirators and Suction Devices

The aspirator (fig. 4, A), known in England as a 'pooter,' is a convenient and effective device for collecting small insects and mites. The following materials are needed to construct an aspirator:

- (1) Vial 2.5-5 cm in diameter and about 12 cm long.
- (2) Two pieces of glass tubing about 7 mm in diameter, one piece about 8 cm long and the other about 13 cm long.
- (3) Rubber stopper with two holes in which the tubing will fit snugly.
- (4) Piece of flexible rubber or plastic tubing about 1 meter long, with diameter just large enough to fit snugly over one end of shorter piece of stiff tubing.

(5) Small piece of cloth mesh, such as cheesecloth, and rubberband.

To make an aspirator, bend the glass tubes as in figure 4, A. In bending or cutting glass tubes, always protect your fingers by holding the glass between several layers of cloth. Obtain the advice of a chemist or laboratory technician for cutting and bending glass. Moisten one end of the longer tube and insert it through one of the holes in the rubber stopper. Moisten one end of the shorter tube, insert it through the other hole in the stopper, and using a rubberband fasten the cloth mesh over the end that was inserted through the stopper; this will prevent specimens from being sucked into the collector's mouth when the aspirator is used. Attach one end of the flexible tubing to the free end of this tube. The length, size, and amount of bend in the tubing will vary according to the user's needs. To complete the assembly, insert the rubber stopper into the vial. To use the aspirator, place the free end of the flexible tubing in the mouth, move the end of the longer glass tube close to a small specimen, and suck sharply. The specimen will be pulled into the vial.

Instead of using a vial, some workers prefer a tube (fig. 4, *B*). In either method, it is well to keep small pieces of absorbent tissue in the vial or tube at all times to prevent moisture from accumulating. Be cautioned that there is some danger of inhaling harmful substances or organisms when using a suction-type aspirator (see ref. 220).

Either the vial- or tubing-type aspirator (fig. 4, *B*) may be converted into a blow-type aspirator by removing the 13-cm glass tube (see fig. 4, *A*) and substituting a T-shaped attachment (fig. 4, *B*). The flexible tubing is attached to one arm of the 'T,' the opposite arm is left open, and the stem of the 'T' is inserted into the vial and covered with mesh. Upon blowing through the flexible tubing, a current of air passes across the 'T' and creates a partial vacuum in the vial, which produces the suction needed to draw specimens into the vial. This kind of aspirator eliminates the danger of inhaling small particles, fungus spores, or noxious fumes.

Aspirators with a squeeze bulb may sometimes be purchased, or if a valved bulb can be obtained, they may be constructed for use with either pressure or suction. Collection traps also have been devised with the suction feature applied on a much larger scale than with the usual aspirator. Suction produced by a fan has been employed in traps in conjunction with light or other attractants. Some of these traps are described in the following references and in the section on Traps. Suction is created by a piston in a 'slurp-gun' described for aquatic collecting. This principle could be adapted for use in air to gather insects and to deposit them in a vial attached to the side of the piston.

References: 11, 20, 48, 84, 120, 142, 220, 226-228, 276, 300, 313, 382, 416, 428, 447, 450, 456, 464.

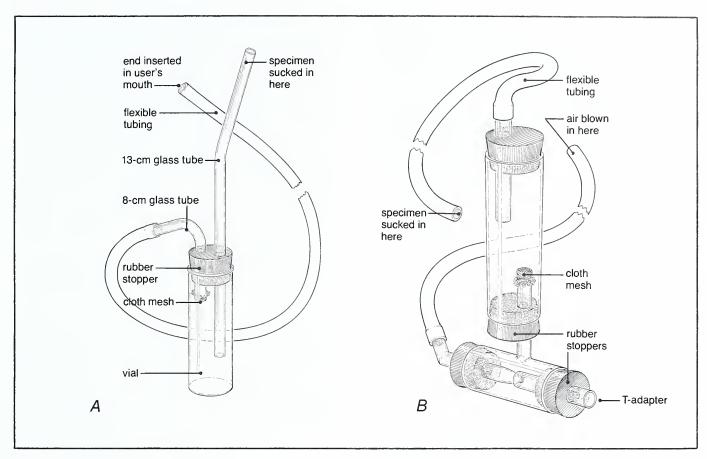


Figure 4. *A*, Assembled vial aspirator; *B*, tube aspirator with blow-type T-adapter.

Beating Sheets

A beating sheet should be made of durable cloth. preferably white, attached to a frame about 1 meter square, with two pieces of doweling or other light wood crossing each other and fitted into pockets at each corner of the cloth (fig. 5). An ordinary light-colored umbrella also may be used as a beating sheet. Place the beating sheet or umbrella under a tree or shrub and sharply beat the branches or foliage with a club or stick. Specimens will fall onto the sheet and may be removed from the light-colored material by hand or with forceps, a moistened brush, or an aspirator. Locating specimens on the sheet is sometimes a problem because of leaves or other unwanted material dropping onto the sheet. Watching for movement will help locate specimens, as well as tilting the sheet so that the debris is displaced or even allowed to fall off, with the insects and mites left clinging to the cloth.

Beating sheets are especially useful in collecting beetles, and they are most useful when the weather has turned cold, or early and late in the day, when normally active insects seek shelter in vegetation and are otherwise difficult to detect.

A 'ground cloth' also is used in sampling crop fields (see ref. 371).

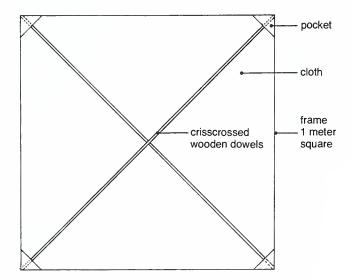


Figure 5. Beating sheet with dowels fastened in corner pockets.

Sifters

Sifters are used to collect insects and mites that live in ground litter, leaf mold, rotting wood, mammal and bird nests, fungi, shore detritus, lichens, mosses, and similar material. Sifters are especially useful for winter collecting to pick up hibernating specimens. Almost any container with a wire-mesh screen bottom will serve as a sifter. The size of the mesh depends on the size of the specimens sought. For general purposes, screening with 2.5-3 meshes per centimeter is satisfactory. To use the sifter, place the material to be sifted into the container and shake it gently over a white pan or piece of white cloth. As the insects and mites fall onto the cloth, they may be collected with forceps, a brush, or an aspirator.

A similar method is used chiefly to collect mites from foliage. Using a sifter of 20-mesh screen (about 8 per centimeter) with a funnel underneath that leads to a small vial, beat pieces of vegetation against the screen to dislodge the mites, which will fall through the screen and into the vial below.

Separators and Extractors

Somewhat similar to the sifter are various devices designed to separate or extract live specimens from substances in which they may be found, such as leaf mold and other kinds of vegetable matter, shore detritus, dung, even net sweepings that include so much foreign matter that it is difficult to pick out the insects. These devices usually depend on some physical aid such as light, heat, or dryness to impel the insects to leave the foreign matter.

One of the simplest such devices is the sweeping separator (fig. 6). This is simply a carton or wooden box with a tight-fitting lid. Near the top of the box on one side is inserted a glass jar. If the jar is made with a screw top, a hole of proper diameter cut in the side of the carton will permit the jar to be screwed onto it. The cover ring, without the lid, from a home-canning jar may be nailed to the periphery of a hole in a wooden box and the jar then screwed onto the ring.

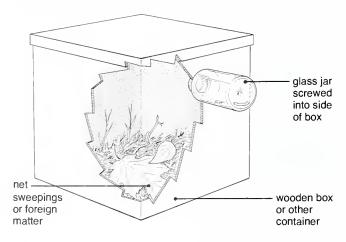


Figure 6. Sweeping separator for extracting live specimens from debris.

The sweepings are dumped into the box and the cover is quickly closed. The insects in the darkened box soon will be attracted to the lighted glass jar. When all the insects appear to have entered the jar, it can be removed and its contents put into a killing jar. Alternatively, a jar cover containing a piece of blotting paper soaked with xylene may be placed over the jar for awhile to stun the insects, which may then be sorted.

The Berlese funnel (fig. 7) and its modifications are cleaner and more efficient than sifting to separate insects and mites from leaf mold and similar materials. The sample is placed on a screen near the top of a funnel. A light bulb can be placed above the sample to produce heat and light, which drive the insects downward into the funnel, or heated coils or a jacket around the funnel can be used to dry the sample and make it inhospitable. The insects and mites are directed by the funnel into a container, sometimes containing alcohol, at the bottom of the funnel. Care should be taken not to dry the sample so rapidly that slow-moving specimens are immobilized before they can leave the sample. To prevent large amounts of debris from falling into the container, place the sample on the screen before the container is put in place.

References: 59, 122, 127, 173, 232-234, 256, 286, 319, 326, 374.

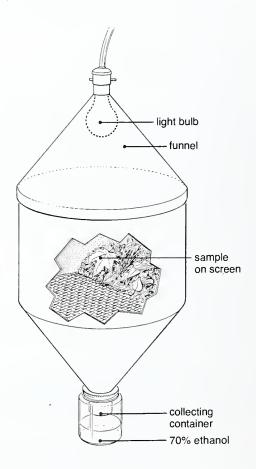


Figure 7. Berlese funnel.

Traps

Since a trap is defined as anything that impedes or stops the progress of an organism, this subject is extensive, including devices used with or without baits, lures, or other attractants. Besides its construction, the performance of a trap depends on such factors as its location, time of year or day, weather, temperature, and kind of attractant used, if any. A little ingenuity coupled with knowledge of the habits of the insects or mites sought will suggest modifications or improvements in nearly any trap or may even suggest new traps.

Only a few of the most useful traps are discussed here, but the following references describe many more, especially references 285, 345, and 394.

References: 1, 14, 16, 17, 40, 54, 55, 109, 121, 130, 133, 150, 154, 164, 176, 178, 179, 182, 186, 189, 193, 195, 206, 213, 236, 237, 285, 297, 312, 345, 347, 394, 395, 417, 423, 446, 449, 454, 464.

Effects of Elevation. One of the external factors affecting the performance of traps, especially light traps, has been specially studied, namely the effect of the elevation (above sea or ground level) at which the trap is placed when in use. The subject is complex, with many variables related to kinds of insects, locality, and so forth, which are discussed in the following references.

References: 42, 66, 91, 137, 152, 153, 157, 298, 369, 402.



Figure 8. Malaise trap used to capture flying insects near a dense forest.

PN-7189

Windowpane Trap. One of the simplest and cheapest traps is a barrier consisting of a windowpane held upright by stakes in the ground or suspended by a line from a tree or from a horizontal line. A trough filled with a liquid killing agent is so placed that insects flying into the pane drop into the trough and drown. They are removed from the liquid, washed with alcohol or other solvent, then preserved in alcohol or dried and pinned. The trap is not recommended for adult Lepidoptera or other insects that may be ruined if collected in fluid

References: 77, 93, 231, 261, 333, 369, 461.

Interception Nets and Barriers. A piece of netting, 1.8 meters or more in height, can be stretched between three trees or poles to form a V-shaped trap with the wide end of the V open. A triangular roof should be adjusted to slope gently downward to the broad open side of the V. A device of this type will intercept many kinds of flying insects, particularly if the trap is situated with the point of the V toward the side of maximum light and in the direction of air movement. A pair of such nets set in opposite directions, or a single net in a zigzag shape, will intercept specimens from two directions. Since insects flying into such a net tend to gather at the pyramidal apex, they are easy to collect. The socalled 'funnel' or 'ramp' traps are interception devices that direct insects to a central point, where a retaining device or killing jar may be placed.

References: 147, 162, 199, 229, 260, 294, 332, 338, 339, 405.

Malaise Traps. It is a small step from the preceding device to the more complex trap developed by the Swedish entomologist René Malaise and that now bears his name. Several modifications of his original design have been published, and at least one is available commercially. The trap, as originally designed, consists of a vertical net serving as a baffle, end nets, and a sloping canopy leading up to a collecting device (fig. 8). The collecting device may be a jar with either a solid or evaporating killing agent or a liquid in which the insects drown. The original design is unidirectional or bidirectional with the baffle in the middle, but more recent types include a nondirectional type with cross baffles and with the collecting device in the center. Malaise traps have been phenomenally successful, sometimes collecting large numbers of species that could not be obtained otherwise. Attractants may be used to increase the efficiency of the traps for special purposes.

References: 64, 405 (bibliography).

Pitfall and Dish Traps. Another simple but very effective and useful type of interception trap consists of a jar, can, or dish sunk in the earth (fig. 9). A cover must be placed over the open top of the jar to exclude rain and small vertebrates while allowing insects and mites to enter. A piece of bark, wood, or flat stone will serve this purpose. Pitfall traps may be baited with various substances, depending on the kind of insects or mites the collector hopes to capture. Although most that fall into the trap will remain there, it should be inspected daily, if possible, and desired specimens removed and placed in alcohol or in a killing bottle while they are in their best condition.

Also in the pitfall category is the cereal dish trap, which is a simple but effective device for obtaining insects attracted to dung. It consists of a small dish, preferably with a rim, set in the earth (fig. 10) and partly filled with 70 percent ethanol, or, if available, with ethylene glycol, which does not evaporate. A piece of stout wire, such as a coathanger, is bent as shown, with a loop at one end to hold the bait receptacle. A few zigzag bends in the other end of the wire will keep the looped end from swinging after the wire is pushed into the earth. The bait receptacle may be a small plastic or metal cup such as is often used for medicine doses, or a coffee creamer, or a cup formed from aluminum foil. When baited with animal or human feces, this trap attracts beetles, mostly of the families Scarabaeidae and Staphylinidae, springtails, ants, earwigs, some parasitic Hymenoptera, and, rather surprisingly, several families of flies, especially Phoridae, Sepsidae, and Muscidae. The larger, strong-flying calliphorid and sarcophagid flies seldom fall into the liquid, although they are attracted to the bait. The alcohol fumes probably cause the smaller flies to drop into it. The trap is made of easily obtained materials, is easily transported, and provides excellent results. It deserves wide use.

References: 5, 17, 28, 51, 103, 126, 149, 155, 166-170, 191, 212, 230, 272, 274, 275, 287, 309 (bibliography), 314, 328, 358, 379, 385, 389, 390, 420, 427, 444.

Emergence and Rearing Traps. An emergence trap is any device that prevents adult insects from dispersing when they emerge from their immature stages in any substrate, such as soil, plant tissue, or water. A simple canopy over an area of soil, over a plant infested with larvae, or over a section of stream or other water area containing immature stages of midges, mayflies, and other arthropods will secure the emerging adults. If it is equipped with a retaining device, as in the Malaise trap, the adults can be killed and preserved shortly after emergence. It must be remembered, however, that many insects should not be killed too soon after emergence because the adults are often teneral or soft bodied and incompletely pigmented and must be kept alive until the body and wings completely harden and colors develop fully. Emergence traps and rearing cages (fig. 11) enable the insects to develop naturally while insuring their capture when they mature or when larvae emerge to pupate.

References: 4, 16, 18, 64, 75, 78, 92, 97, 100, 104, 145, 151, 187, 207, 236, 247, 250, 253, 257, 263, 267, 277, 278, 290, 295, 307, 310, 315-318, 320, 325, 331, 390, 422, 429, 469.

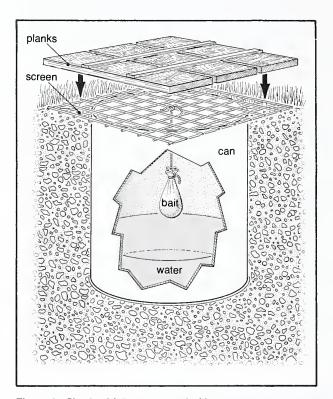


Figure 9. Simple pitfall trap covered with screening and provided with hanging bait.

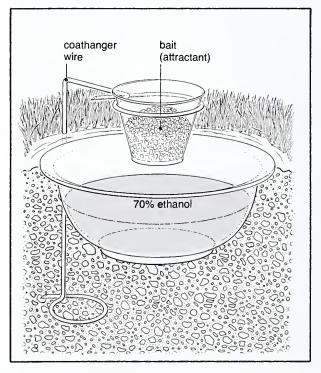


Figure 10. Cereal dish trap.

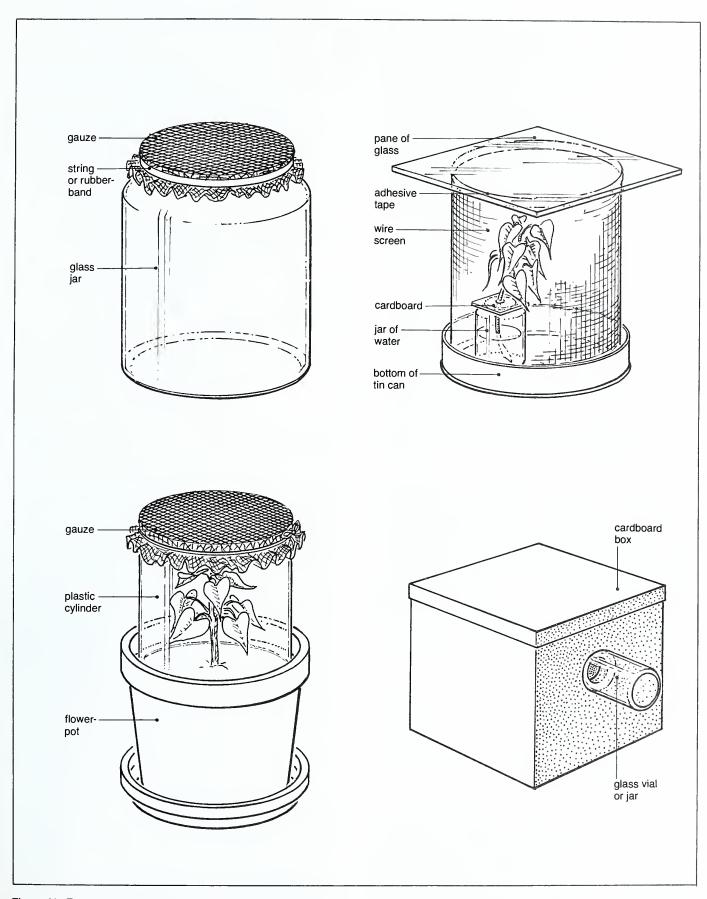


Figure 11. Emergence traps and rearing cages, with living plant material in two containers.

Traps Using the Lobster or Eel Trap Principle. This category includes any container that has its open end fitted with a truncated cone directed inward, as in a lobster or eel trap, known as a 'Reuse' in German. An ordinary killing jar with a funnel fastened into its open end is an example. When the funnel is placed over an insect, the specimen will usually crawl or fly toward the light and enter the jar through the funnel. Modified traps of this type include the Steiner and McPhail traps, which are used primarily in fruit fly surveys but are suitable for many other purposes. The inside of the Steiner trap usually has a sticky material containing a pheromone or other lure. Both traps, as well as similar devices, may be used with different attractants to collect diverse kinds of insects.

References: 31, 56, 57, 104, 207, 223, 310, 322, 329, 331, 361, 404.

Light Traps. With light traps, advantage is taken of the attraction of many insects to a light source. Using various wavelengths as the attractant, a great variety of traps can be devised, a few of which are described here.

Many traps can be constructed easily from materials generally available around the home. All wiring and electrical connections should be approved for outdoor use. Funnels can be made of metal, plastic, or heavy paper. Traps can be used with or without a cover, but if they are to be operated for several nights, covers should be installed to keep out rain.

The New Jersey trap (fig. 12) includes a motorized fan to force insects attracted to the light into a killing jar. It has been especially useful for collecting small, non-scaly insects such as midges and gnats. This type of light trap, in which the insects fall directly into a killing jar, is not recommended for use with moths because such delicate specimens may be badly rubbed or torn. If only small insects are desired, they may be protected from damage by larger insects by placing a screen with the proper sized mesh over the entrance. The Minnesota trap is very similar to the New Jersey trap, but it does not include a fan or any motorized method of draft induction.

The Wilkinson trap (fig. 13) requires somewhat more effort to construct than the preceding traps, but it has the advantage of confining, not killing, the trapped insects. Moths, therefore, can be collected in good condition if the trap is inspected frequently and desirable specimens are removed quickly through the hinged top and placed in a killing jar.

Several highly effective but more elaborate devices have been made for collecting moths and other fragile insects in good condition. Basically, they all use the principle of a funnel with a central light source above it and vanes or baffles to intercept the approaching insects that are dropped through the funnel into the container beneath, which may or may not hold a killing agent. The nature of the container and the type of kill-

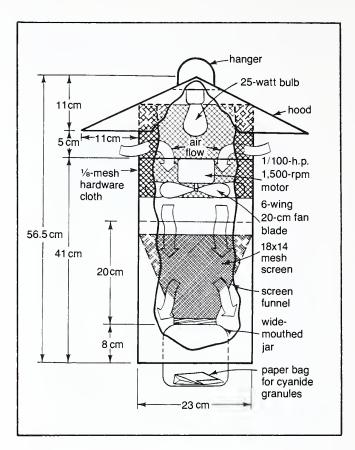


Figure 12. New Jersey trap.

ing agent affect the quality of the specimens obtained. Some traps catch the insects alive in a large collection chamber, such as a garbage can, which is filled or nearly filled with loosely arranged egg cartons. Most moths will come to rest in the cavities between the egg cartons and will remain there until removed in the morning.

Other traps are designed to kill the insects by means of high concentrations of fumes from a liquid killing agent, such as tetrachloroethane or calcium cyanide (the "cyanogas" of the exterminating industry). A heaping tablespoon or more of calcium cyanide is placed in each of four to six brown paper bags, which are hung in a large garbage can or other large container. A dampened cloth, such as a washcloth, is also hung inside the can to humidify the air and activate the cyanide. This is especially necessary in dry weather. The concentration of the gas inside the can is so great that insects are inactivated almost instantly on entering, and even the most delicate specimens are damaged very little. The bags containing the calcium cyanide powder should be replaced as needed. If two of the oldest bags are replaced with two fresh ones each successive night, the trap can be run as long as the collector desires.

Handle cyanide outdoors, facing downwind, and with extreme caution. During the day, when the trap is not in use, store the cyanide bags in an airtight container. All forms of cyanide used as killing agents react and break

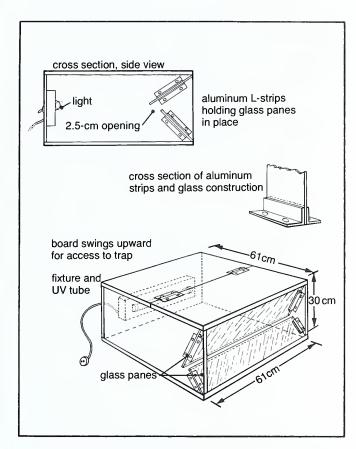


Figure 13. Wilkinson trap.

down quickly when exposed to air and moisture; nevertheless dispose of the residue carefully.

To prevent rainwater from accumulating in the trap, place a screen-covered funnel inside the collection chamber to drain the water out through a hole in the bottom of the trap. Sometimes a system of separators is added to guide beetles and other heavy, hard-bodied insects into a different part of the container than the moths and other delicate specimens.

The most efficient light traps use lamps rich in their output of ultraviolet light. The British-made Robinson trap employs an intense, blue-white, 125-watt mercury vapor lamp of a type used for street lighting. This, the most effective insect attractant commercially marketed, is widely used in many kinds of light traps because it has some special advantages over other kinds of attractants. For example, this type of lamp is the only one that emits the kind of light that attracts large numbers of *Catocala* (underwing) moths, a colorful group popular with many collectors.

Many traps are equipped with 15-watt ultraviolet fluorescent tubes, which emit a highly visible bluish-white light, although blacklight tubes emitting deep purple light are similarly effective. Ultraviolet tubes of lower or higher wattage also may be used and are all highly effective. A 15-watt ultraviolet tube has been estimated to attract about 10 times as many insects as a 500-

candlepower gasoline lantern or incandescent lamp. The advantage of the fluorescent tube over the mercury vapor lamp is that it is less expensive and much more portable. A 15-watt tube is easily powered by an ordinary automobile battery by using an inverter to change 6- or 12-volt direct current to 120-volt alternating current. Also, its ultraviolet output is not strong enough to cause any significant eye damage. The safety factor of the mercury vapor lamp at close range is less certain, although entomologists who have used the Robinson trap for many years seem to have suffered no ill effects.

A new, lightweight, spillproof 12-volt battery, in which the acid electrolyte is a gel rather than a liquid, is far superior to the standard automotive battery for powering light traps, but it is fairly expensive and requires a special charger. Special lightweight, nickel-cadmium battery packs, used to power blacklights for collecting, are marketed by some dealers of entomological equipment.

Light Sheets. Another highly effective method of using light to attract moths and other nocturnal insects is with a light sheet. This is simply a cloth sheet, usually a white bedsheet, hung outdoors at night with an appropriate light source or combination of sources such as ultraviolet fluorescent tubes, gasoline lanterns, or automobile headlights placed a few feet in front of it. As insects are attracted and alight on the sheet, they are easily captured in cyanide bottles or jars by the collector who stands in attendance or at least checks the sheet frequently. The sheet may be pinned to a rope tied between two trees or fastened to the side of a building, with the bottom edge spread out on the ground beneath the light. Some collectors use supports to hold the bottom edge of the sheet several centimeters above the ground so that no specimens can crawl into the vegetation under the sheet and be overlooked. Other collectors turn up the edge to form a trough into which insects may fall as they strike the sheet.

The light sheet remains unsurpassed as a method of collecting moths in flawless condition or of obtaining live females for rearing purposes. Its main disadvantage is that species that fly very late or those that are active only in the early morning hours may be missed unless one is prepared to spend most of the night at the sheet. Many other insects besides moths are attracted to the sheet, and collectors of beetles, flies, and other kinds of insects would do well to collect with this method.

It should be emphasized that the phases of the moon most profoundly influence the attraction of insects to artificial light. Attraction is inhibited by a bright moon. The best collecting period each month extends from the fifth night after the full moon until about a week before the next full moon.

References (light traps and sheets): 7, 8, 23-25, 33, 34, 42, 50, 61, 73, 74, 81, 99, 101, 135, 136, 139, 162, 174, 181,

189, 204, 205, 213, 244, 273, 281, 298, 299, 308, 313, 323, 336, 350, 351, 391, 396, 401, 402, 418, 430, 447, 451, 453.

Color Traps. Colored objects are also used as attractants for insects. A bright yellow pan containing water to which a little detergent has been added to reduce surface tension and thereby cause insects to drown more quickly is used to collect winged aphids. The insects are attracted by the color and drown in the water. Yellow seems to be the best color for traps, but various kinds of insects react differently to different colors. The Manitoba trap (fig. 14) has a black sphere to attract horse flies (family Tabanidae), which are then captured in a canopy-type trap.

References: 36, 164, 174, 211, 235, 246, 355.

Sticky Traps. In this type of trap, a board, piece of tape, pane of glass, piece of wire net, cylinder, or other object, often painted yellow, is coated with a sticky substance and suspended from a tree branch or other convenient object. Insects landing on the sticky surface are unable to extricate themselves. The sticky material is later dissolved with a suitable solvent, usually toluene, xylene, ethylacetate, or various combinations of these, and the insects are washed first in Cellosolve and then in xylene (see p. 25). This type of trap should not be used to collect certain specimens, such as Lepidoptera, which are ruined by the sticky substance and cannot be removed without being destroyed.

Various sticky-trap materials are available commercially, some with added attractants. However, use caution in selecting a sticky substance because some are difficult to dissolve.

References: 62, 79, 107, 112, 121, 148, 155, 156, 158, 183, 185, 192, 226, 252, 289, 292, 305, 319 (pp. 226-227), 319a, 354, 406, 417, 456.

Snap Traps. Two kinds of traps designed for quantitative sampling may be termed "snap traps." One of them (see ref. 293) consists of a pair of wooden or plastic discs, slotted to the center so as to fit on a tree branch and connected to each other by a pair of rods. A cloth cylinder is affixed at one end to one of the discs and at the other end to a ring sliding on the rods. After the cloth cylinder has been pulled to one end and has been secured in place, the ring is held by a pair of latches. When insects have settled on the branch, its leaves, or flowers, the latches are released by pulling on a string from a distance, and the trap is snapped shut by a pair of springs on the rods, capturing any insects present. One of the canopy traps (see ref. 428) operates in a similar fashion. When a remotely controlled latch is pulled, a spring-loaded canopy is snapped over an area of soil, and insects within the canopy are collected by suction or a vacuum device. This trap was designed for use in grasslands.

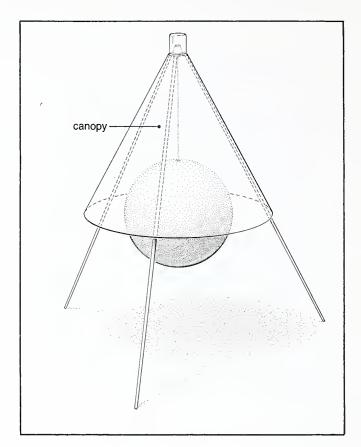


Figure 14. Manitoba trap for collecting horse flies.

Artificial Refuges. Many insects, especially beetles, are successfully found under stones, planks, or rotten logs. Providing such refuges, as pieces of wood, cardboard, or even complex traps, is also a form of trapping.

References: 67, 385.

Electrical Grid Traps. In recent years, electrocuting pest insects has been used extensively in control work. The insects are attracted to a device by a pheromone or other lure placed in a chamber protected by a strongly charged electrical grid. The method deserves study for other purposes, such as surveying the arthropod fauna of an area.

References: 158, 186, 301-303, 366, 397.

Baits, Lures, and Other Attractants

Any substance that attracts insects may be used as a bait. Natural products, chemicals derived therefrom or synthesized, and secretions of the insects themselves may all be used as attractants. Mere exposure of the substance may be considered as setting up a trap, and attractive substances are used in many constructed traps.

Sugaring for moths, one of the oldest collecting methods, involves the use of a specially prepared bait in which some form of sugar is an essential component.

The bait may be refined or brown sugar, molasses, or sirup. Such substances often are mixed with stale beer, fermented peaches, bananas, or some other fruit—there is no standard formula. Each lepidopterist has his or her own favorite recipe.

One particularly satisfactory recipe uses fresh, ripe peaches; culls or windfalls are suitable. Remove the seeds but not the skins, mash the fruit, then place it in a 4-liter (1-gal) or larger container of plastic, glass. stainless steel, enamelware, or crockery with a snugly fitting but not tight cover. Avoid using metal containers that may rust or corrode. Fill each container only onehalf to two-thirds full to allow space for expansion. Add about a cup of sugar and place in a moderately warm place for the mixture to ferment. The bubbling fermentation reaction should start in a day or so and may continue for 2 weeks or more, depending on the temperature. During this time, check the fermentation every day or every other day and add sugar until fermentation appears to have subsided completely. As the added sugar is converted to alcohol, the growth of yeast slows and eventually ceases.

After fermentation ceases, the bait should remain stable and should then be kept in tightly sealed containers to prevent contamination and evaporation. If the mixture is allowed to run low in sugar during the fermentation process, vinegar will be produced instead of alcohol. It is therefore important to smell the bait periodically and to add plenty of sugar to avoid this. The amount of sugar consumed will be surprising, usually over 0.4 kg per liter (3.3 lb per gal). The bait should have a sweet, fruity, winelike fragrance. A trace of vinegar is not objectionable but is better avoided. Canned fruit, such as applesauce, may also be used to make the bait, but inasmuch as such products are completely sterile, a small amount of yeast must be added to start fermentation. Although the bait may seem troublesome to prepare, it keeps for years and is thus available at any time, even when fruit is not in season.

Immediately before use, the bait may be mixed with 30 to 50 percent molasses or brown sugar or a mixture of these. This thickens the bait so that it will not dry out so quickly, and it makes the supply last longer.

The best time to set out the sugar bait is in the early evening before dark. It may be applied with a paint brush in streaks on tree trunks, fenceposts, or other surfaces. Choose a definite route, such as along a trail or along the edge of a field, so that later you can follow it in the dark with a lantern or flashlight. Experienced collectors learn to approach the patches of bait stealthily with a light in one hand and a killing jar in the other to catch the moths before they are frightened off. Some collectors prefer to wear a headlamp, leaving both hands free. Although some moths will fly away and be lost, a net usually is regarded as an unnecessary encumbrance, because moths can be directed rather easily into the jar. Sugaring is an especially useful way to collect noctuid moths, and the bait applied in the even-

ing often will attract various diurnal insects on the following days. The peach bait previously described has been used in butterfly traps with spectacular results. However, collecting with baits is notoriously unpredictable, being extremely productive on one occasion and disappointing on another, under apparently identical conditions.

Baiting With Feces. Animal and human feces attract many insects. A simple but effective method of collecting such insects is to place fresh feces on a piece of paper on the ground and wait a few minutes. When a sufficient number of insects have arrived, a net with its bag held upward can be brought carefully over the bait about 1 meter above it. This will not disturb the insects, nor will they be greatly disturbed when the net is lowered gently about two-thirds of the distance to the bait. At this point, the net should be quickly lowered until its rim strikes the paper. The insects, mostly flies, will rise into the net, which may then be lifted a short distance above the bait and quickly swung sideways, capturing the insects in the bottom of the bag. In about half an hour, many flies can be caught, virtually all that have come to the bait. Because of this, the 'baiting with feces' method may be used for quantitative studies (see ref. 403).

Feces are by far most attractive to insects during the first hour after deposition, but insects coming for a more extended period may be captured by placing a canopy trap over the feces or by using the feces with the cereal dish trap (see p. 12). Emergence traps placed over old feces will capture adult insects emerging from immature forms feeding there. The same methods also may be used with other baits, such as decaying fruit, small carcasses, and a wide variety of other substances.

The Oatmeal Trail. Hubbell (ref. 218) showed that dry oatmeal scattered along a path will attract such insects as crickets, camel crickets, cockroaches, and ants. Some of these insects feed only at night and may be hand-collected by flashlight or by light from a head-lamp. A killing bottle is used, and the specimens are collected with fingers, an aspirator, or a net.

Pheromones and Other Attractants. Substances naturally produced by insects to attract others of their own kind are known as pheromones. They are often used in traps to aid in controlling pest species. Most pheromones are highly specific, attracting only one species or a group of closely related species. "Spanish Fly" (cantharidin) has recently come into use as an extremely effective attractant for various beetles, such as pedilids, and bugs, such as bryocerines. Female specimens of certain insects, such as cicadas and silkworm moths, may be placed alive in a trap and used as a bait with their pheromones and the sounds they produce attracting males. A female saturniid (silkworm moth) will attract males from a great distance.

Host animals likewise may be used as bait for various bloodsucking insects, with or without constructed traps. Carbon dioxide in the form of "Dry Ice," cylinder gas, or marble chips treated with an acid such as vinegar serves as an attractant for certain insects and has been very successful in attracting horse flies to Malaise and Manitoba traps. Sounds are produced by many insects to attract others of their own kind. These sounds are very specific in pitch, tempo, and duration. Recordings of such sounds, played at the proper volume, have been effective in luring grasshoppers, crickets, and other kinds of insects.

References for attractants: General—2, 10, 29, 35-37, 49, 72, 85, 86, 100, 102, 123, 155, 168, 198, 213, 218, 223, 251, 259, 262, 275, 283, 288, 311, 321, 327, 349, 362, 365, 376, 384, 403, 408, 438, 445, 462; carbon dioxide—44, 72, 97, 100, 121, 123, 148, 217, 231, 239, 311, 327, 359, 360, 362, 365, 392, 409, 413, 448, 460; pheromones—28, 31, 38, 41, 67, 68, 159, 189, 203, 213, 216, 222, 223, 301, 324, 340, 383, 384, 395, 398, 441; sound—32, 65.

Collecting Aquatic and Soil Insects and Ectoparasites

Insects and mites emerging from water may be collected by the same means as terrestrial insects, but specialized equipment is required, which is not described here, since aquatic insects are of relatively little direct importance to agriculture. However, they are of great importance in public health and general ecological studies. The following references pertain to aquatic collecting.

References: 8, 73, 92, 94, 110, 111, 114, 118, 145, 200, 229, 236, 250, 257, 258, 262, 277, 278, 289, 290, 296 (general), 307, 315-318, 320, 341 (general), 348, 388, 414, 415, 439, 443 (general), 465, 466.

As with aquatic specimens, insects and mites that live on or under the soil surface require special techniques and equipment for their collection and study. Many soil-inhabiting species are of great economic importance because they devour the roots of crops. Many spend their immature stages in soil but emerge and leave the soil as adults. A considerable amount of literature on soil insects has been published, the most useful of which is cited here. See also the references cited under Separators and Extractors (p. 10) and Pitfall and Dish Traps (p. 12).

References: 21, 51, 52, 103, 125, 233, 234, 249, 256, 282, 319, 326, 375, 419.

Some ectoparasites, particularly those that fly, may be collected in some of the traps discussed (p. 17), using their hosts as bait; others may be collected by means of the special devices described in the following references.

References: 53 (p. 152), 89, 440, 458.

Rearing

Collectors should take every opportunity to rear insects and mites, for not only are reared specimens generally in the best possible condition, but rearing provides life stages that otherwise might be collected only rarely or with great difficulty. By preserving one or more specimens from each of the stages as they are reared, if sufficient material is available, the collector can obtain series of immature stages along with associated adults. Such series are desirable, especially for species in which the adult is known but the immature stages are unknown or difficult to identify. The converse often is true also-some species of insects, such as stem-mining flies, are fairly abundant in the larval stage but have never been reared to the adult stage; consequently, one does not know whether they are stages of a species that has been described and named from an adult but whose life history is unknown. Since adults of these flies are seldom found, the easiest way to obtain the stage necessary for specific determination is to rear the larvae or pupae.

If only a few specimens are reared, the shed skins and pupal cases or puparia should be preserved, as they are of value if properly associated with the reared adult. Do not preserve a pupa or puparium with an adult unless you are positive that the association is correct. It is best to put pupae in separate containers so that adults or parasites that emerge are associated with certainty. If at all feasible, the parasite's host should be preserved for identification. Keep careful notes throughout the rearing so that all data relative to the biology of the species are properly correlated.

Containers for Rearing

To rear specimens successfully, simulate as closely as possible in the rearing cages the natural conditions under which the immatures were found. Almost any container will serve as a temporary cage for living insects or mites. One simple temporary cage that is very handy on field trips is a paper bag. Plant material or a soil sample containing insects or mites is placed in the paper bag, which is then sealed. A paper bag also can be placed over the top of a plant on which insects or mites are found. The bottom edge of the bag is tied tightly around the exposed stems, which are cut and placed in a jar of water. One disadvantage of using a paper bag is that it is not transparent, so it must be removed to observe the specimens or to determine when the foliage needs to be changed. Clear plastic bags are better suited to such viewing; however, they are not recommended for more than short-term use because they are airtight, and specimens may be ruined by drowning in condensed water on the inside of the

Another simple temporary cage is a glass jar with its lid replaced by a piece of organdy cloth or gauze held in place by a rubberband. A few such jars in a collecting kit are useful for holding live insects. For aquatic

species, using a watertight lid on the jars is advisable. If aquatic insects are to be transported over a considerable distance, fewer will die if the jar is packed with wet moss or leaves than if the specimens are allowed to slosh around in water alone. After arrival at your destination, release the insects into a good rearing container (fig. 11).

Aquatic insects can be reared in their natural habitat by confining them in a wire screen or gauze cage, part of which is submerged in water. Be sure to anchor the cage securely. The screen used in aquatic cages should be coarse enough to allow food to flow through, yet fine enough to retain the insects being reared. Certain aquatic insects may be reared readily indoors in an aquarium or even in a glass jar. The main goal is to try to duplicate their natural habitat. If the specimen was collected from a rapidly flowing stream, it is unlikely to survive indoors unless the water is aerated. Other insects do well in stagnant water. Aquatic vegetation usually should be provided in the aquarium even for predaceous specimens, such as dragonfly nymphs, which often are found clinging to underwater stems. Keep sufficient space, which will vary according to the insect being reared, between the surface of the water and the screen or gauze cover over the aquarium to allow the adult insect to emerge. A dragonfly, for example, needs considerable space, plus a stick, rock, or other object on which to perch after emerging so that the wings will develop fully.

Most adult insects, both terrestrial and aquatic, are teneral when they first emerge and should not be killed until the exoskeleton and wings harden and the colors develop fully. This may be a matter of minutes, hours, or even days. It is advisable to keep even small flies alive for 1 full day after they emerge. Specimens killed while still teneral will shrivel when mounted. Some insects, if kept in cages too long after emerging, especially butterflies and moths, will beat their wings against the cage and lose many scales or tear their wings. Providing adequate space in which emerging insects may expand their wings fully and move about slightly is therefore critical in the design of rearing cages.

Beetles and other boring insects often are abundant in bark and wood. If pieces of such material are placed in glass or metal containers, excellent specimens of the adults may be obtained, although sometimes not for a considerable time. Cages made of wood or cardboard are not suitable for such insects because those found in wood or bark usually are well equipped, both in immature and adult stages, to chew their way through a cage made of such material and thus escape.

A flowerpot cage is one of the best containers for rearing plant-feeding species over an extended period. The host plant, if its size and habitat permit, is placed in a flowerpot, and a cylinder of glass, plastic, or wire screen is placed around the plant (fig. 11, lower left).

Another type of flowerpot cage is made by inserting a cane or stick, taller than the plant, into the soil in the pot. One end of a net or muslin tube is fitted over the edge of the pot and is held in place by a string. The other end of the tube is tied around the top of the stick. An advantage of the flowerpot cage is that the plant is living, and fresh plant material need not be added daily.

Plant-feeding mites will not wander far as long as suitable host material is available for them. Because mites are wingless even as adults, they can be confined in an open rearing container by making a barrier around the top edge or upper inner sides of the container with Vaseline or talcum powder.

Emergence cages are essentially rearing cages that are used when it is impractical or impossible to bring specimens indoors. Emergence cages may also be considered as traps and are discussed under that heading (see p. 12). With plant-feeding insects, a sleeve consisting of a muslin tube with open ends is slipped over a branch or plant and tied at one end. The insects are then placed in the tube, and the loose end of the tube is tied. This cloth tube can be modified to allow observation of the insects by replacing the midsection with a "window" of clear plastic or wire screen. If the insects in the tube require duff or debris in which to pupate, the tube should be placed perpendicular to the ground and duff or debris placed in the lower end.

Rearing Conditions and Problems

Moisture. The moisture requirements of insects and mites are varied. Examination of the habitat from which specimens were collected should provide clues about their moisture requirements in captivity. Many insects in the pupal stage are resistant to drought. Species that normally infest stored foods also require very little moisture; in fact, many produce their own water. Most species found outdoors require higher levels of humidity than are generally found indoors. Additional moisture can be added to indoor rearing cages in several ways. To increase the humidity in a cage, keep a moist pad of cotton on top of the screen cover of the cage, or place a moist sponge or a small glass vial filled with water in the cage. The mouth of the vial is plugged with cotton and the vial laid on its side so the cotton remains moist. Pupae may be held for long periods in moist sawdust, vermiculite, sphagnum, or peat moss. In a flowerpot cage, the water used to keep the plant alive should provide sufficient moisture for the plant-feeding insects and mites. Spraying the leaves daily also may supplement moisture requirements in rearing cages. Too much moisture may result in water condensation on the sides of the cage, which may trap the specimens and damage or kill them. Excess moisture also enhances the growth of mold and fungus, which is detrimental to the development of most insects and mites. A 2-3 percent solution of table salt sprayed regularly in the cage will help prevent mold and fungus growth.

Temperature. Of all the environmental factors affecting the development and behavior of insects and mites, temperature may be the most critical. Since arthropods are cold blooded, their body temperatures are usually close to the temperature of the surrounding environment, and their metabolism and development are directly affected by increases and decreases in temperature. Each stage of an insect or mite species has a low and a high point at which development ceases. These are called threshold temperature levels.

Most species that are collected and brought indoors for rearing can be held at normal room temperature; the optimum temperature for rearing will vary from species to species and with different stages of the same species. As with all rearing techniques, every attempt should be made to duplicate natural conditions. Specimens that normally would overwinter outdoors should be kept during the winter in rearing cages placed in an unheated room, porch, or garage. Never place an enclosed rearing cage in direct sunlight; the heat becomes too intense and may kill the specimens.

Dormancy and Diapause. Insects and mites are unable to control the temperature of their environment; instead, they make physiological adjustments that allow them to survive temperature extremes. In regions with freezing winters, insects and mites have at least one stage that is resistant to low temperatures. The resistant form may be any stage-egg, larva, nymph, pupa, or adult. When winter arrives, only the resistant form survives. Dormancy is the physiological state of an insect or mite during a period of arrested development, whereas diapause is the prolonged period of arrested development brought about by such adverse conditions as heat, drought, or cold. This condition can be used to advantage in rearing. For example, if leaving rearing cages unattended for several days or longer is unavoidable, many (but unfortunately not all) specimens can be refrigerated temporarily to slow their activity and perhaps force diapause. This measure should be used with caution since the degree and duration of cold tolerated by different species will vary.

The reverse situation, that of causing diapause to end, is equally useful. Overwintering pupae that normally would not develop into adults until spring can be forced to terminate diapause early by chilling them for several weeks or months, then bringing them to room temperature so normal activity will resume. Often mantid egg cases are brought indoors accidentally with Christmas greenery. The eggs, already chilled for several months, hatch when kept at room temperature, often to the complete surprise and consternation of the unsuspecting homeowner.

Light. Most species of insects and mites can be reared under ordinary lighting conditions; however, artificial manipulation of the light period will control diapause in many species. If the light requirements of the species being reared are known, it may be possible

to adjust the period of light so that the specimens will continue to develop and will remain active instead of entering diapause, for example, providing 8-10 hours of light as opposed to 16 hours. Light and dark periods can be regulated with a 24-hour timing switch or clock timer. The timer is set to regulate light and dark periods to correspond with the desired lengths of light and darkness. It is important to remember that many insects and mites are very sensitive to light; sometimes even a slight disturbance of the photoperiod can disrupt their development.

Food. The choice of food depends on the species being reared. Some species are general feeders and will accept a wide assortment of food, including dead or decaying organic matter. Examples of general feeders are most ants, crickets, and cockroaches. Other groups are specific feeders, with food preferences so restricted that only a single species of plant or animal is acceptable. Carefully note at the time of collection the food being consumed by the specimen, and provide the same food in the rearing cages.

Carnivorous insects should be given prey similar to that which they normally would consume. This diet can be supplemented when necessary with such insects as mosquito larvae, wax moth larvae, mealworms, maggots, or other insects that are easily reared in large numbers in captivity. If no live food is available, a carnivorous insect sometimes may be tempted to accept a piece of raw meat dangled from a thread. Once the insect has grasped the meat, the thread can be gently withdrawn. The size of the food offered depends on the size of the insect being fed. If the offering is too large, the feeder may be frightened away. Bloodsucking species can be kept in captivity by allowing them to take blood from a rat, mouse, rabbit, or guinea pig. A human should be used as a blood source only if it is definitely known that the insect or mite being fed is free of diseases that may be transmitted to the human.

Stored-product insects and mites are easily kept alive and breeding in containers with flour, grains, tobacco, oatmeal or other cereal foods, and similar products. Unless leaf-feeding insects are kept in flowerpot cages where the host plant is growing, fresh leaves from the host plant must usually be placed in the rearing cage daily and old leaves removed.

Artificial Diets. Some species can be maintained on an artificial diet. The development of suitable artificial diets is complex, involving several factors besides the mere nutritional value of the dietary ingredients. Because most species of insects and mites have very specific dietary requirements, information regarding artificial diets is found mainly in reports of studies on specific insects or mites.

Special Problems and Precautions in Rearing. Problems may arise in any rearing program. Cannibalism, for instance, is a serious problem in rearing preda-

ceous insects and necessitates rearing specimens in individual containers. Some species resort to cannibalism only if their cages become badly overcrowded. Disease is also a problem. It can be caused by introducing an unhealthy specimen into a colony, poor sanitary conditions, lack of food, or overcrowding.

Cages should be cleaned frequently and all dead or clearly unhealthy specimens removed. Use care not to injure specimens when transferring them to fresh food or when cleaning the cages. Mites and small insects can be transferred with a camel's hair brush.

Attacks by parasites and predators also can be devastating to a rearing program. Carefully examine the host material when it is brought indoors and before it is placed in the rearing containers to lessen the possibility of predators and parasites being introduced accidentally. Also, place rearing cages where they will be safe from ants, mice, the family cat, and other predators.

References (rearing): 16, 18, 83, 128, 141, 144, 187, 200, 247, 263, 296, 345, 388, 431.

Liquid Agents for Killing and Preserving

Insects and mites of all kinds may be killed and preserved in liquid agents, but it is first necessary to determine the advisability of using a liquid killing agent rather than a dry gaseous agent. Some kinds of insects are best kept dry; it may not be advisable to allow others to become dry. Directions for the treatment of various insects are given in the last part of this publication under the various orders.

Preservation of insects in alcohol is a complex subject about which there is a certain amount of controversy and misunderstanding. If one specializes in an insect group suited to preservation in one or another kind or concentration of alcohol, consult specialists in that group or experiment to find what yields the best results.

Ethanol (grain or ethyl alcohol) mixed with water (70 to 80 percent alcohol) is usually the best general killing and preserving agent. For some kinds of insects and mites, other preservatives or higher or lower concentrations of alcohol may be better. Because pure ethanol is often difficult to obtain, some collectors use isopropanol (isopropyl alcohol) with generally satisfactory results. Isopropanol does not seem to harden specimens as much as ethanol, and at least it is satisfactory in an emergency. Although there is controversy over the relative merits of ethanol and isopropanol, the choice of which to use is not so important as what concentration to use. This choice depends on the kind of insect or mite to be preserved.

Parasitic Hymenoptera are best killed and preserved in 95 percent alcohol. This high concentration prevents the membranous wings from becoming twisted and

folded, hairs from matting, and soft body parts from shriveling. This concentration may also be desirable if large numbers of insects are to be killed in a single container, such as in the killing jar of a Malaise trap, because the insect body fluids will dilute the alcohol. On the other hand, soft-bodied insects, such as aphids and thrips, small flies, and mites, become stiff and distorted if preserved in 95 percent alcohol and should be preserved in alcohol of a lower concentration. Adult bees should not be collected in alcohol because their usually abundant body hairs become badly matted. Adult moths, butterflies, mosquitoes, moth flies, and other groups with scales and long, fine hairs on the wings or body may be worthless if collected in alcohol regardless of the concentration.

Formalin (formaldehyde) solutions should not be used because the tissues become excessively hardened and the specimens then become difficult to handle.

Larvae of most insects should be collected in alcohol and subsequently killed in boiling water to "fix" their proteins and prevent them from turning black. Larvae should be left in hot water for 1-5 minutes, depending on the size of the specimens, then transferred to 70-80 percent alcohol. Large specimens or small specimens that have been crowded into one vial should be transferred to fresh alcohol within a day or two to reduce the danger of diluting the alcohol with body fluids. If the alcohol becomes too diluted, the specimens will begin to decompose. Water is not a preservative.

For some groups, preservation is better if certain substances are added to the alcohol solution. Thrips and most mites, for example, are best collected in an alcohol-glycerin-acetic acid (AGA) solution, and for many larvae a kerosene-acetic acid-dioxane (KAAD) solution is preferred. If KAAD is used, larvae need not be killed in boiling water. Formulas for these and other solutions are given in the Appendix.

For some histological, cytological, or physiological studies, specimens must be in a certain critical condition and must be preserved in special media (see ref. 437).

Larvae and most soft-bodied adult insects and mites can be kept almost indefinitely in liquid preservatives; however, for a permanent collection, mites, aphids, thrips, whiteflies, fleas, and lice usually are mounted on microscope slides (see p. 34). Larvae are usually kept permanently in alcohol, but some may be mounted by the freeze-drying technique (see p. 34) or by inflation (see p. 34). Many insects collected in alcohol are later pinned for placement in a permanent collection. Hardbodied insects such as beetles can be pinned directly after removal from alcohol, but for them and all softer insects such as flies and wasps, follow the process described on page 25.

Temporary Storage of Specimens

After specimens have been collected, time is often not immediately available to prepare them for permanent storage. There are several ways to keep them in good condition until they can be prepared properly. The method used depends largely on the length of time that the specimens may have to be stored temporarily.

Refrigeration

Medium to large specimens may be left in tightly closed bottles for several days in a refrigerator and still remain in good condition for pinning as will smaller specimens if left overnight. Some moisture must be present in the containers so that the specimens do not become "freeze-dried," but if there is too much moisture, it will condense on the inside of the bottle as soon as it becomes chilled. Absorbent paper placed between the jar and the insects will keep them dry. When specimens are removed for further treatment, place them immediately on absorbent paper to prevent moisture from condensing on them.

Insects may be placed in alcohol, as described on page 21, and kept for several years before they are pinned or otherwise treated.

Dry Preservation

It is standard practice to place many kinds of insects in small boxes, paper tubes, triangles, or envelopes for an indefinite period, allowing them to become dry. It is not advisable to store soft-bodied insects by such methods because they become badly shriveled and very subject to breakage. Diptera should never be dried in this manner because the head, legs, and most of all the antennae become detached very easily.

Almost any kind of container may be used for dry storage; however, tightly closed, impervious containers of metal, glass, or plastic should be avoided because mold may develop on specimens if even a small amount of moisture is entrapped. Nothing can be done to restore a moldy specimen.

Dry-stored specimens must be labeled with complete collection data in or on each container (see p. 40). Avoid placing specimens collected at different times or places in the same container. If specimens with different collection data must be layered in the same container, include a separate data slip with each layer.

To insure that specimens do not slip from one layer to another, cut pieces of absorbent tissue, glazed cotton, or cellucotton a little larger than the inside of the container. Place a few layers of this material in the bottom of the container, then a few insects (do not crowd them), then more layering material, and so on until the container finally is filled. If much space is left, use a little plain cotton, enough to keep the insects from moving about but not enough to produce pressure that will damage them. To prevent parts of the insects from getting caught in the loose fibers, use plain cotton only for the final layer. Insect parts are very difficult to extract from plain cotton without damage.

One method of keeping layered specimens soft and pliable for several months includes the use of chlorocresol in the bottom of the layered container and a damp piece of blotting paper in the top. The container must be impermeable and sealed while stored; plastic sandwich boxes make useful containers to use with this method. Add about a teaspoonful of chlorocresol crystals to the bottom, cover with a layer of absorbent tissue, follow with the layers of specimens, then a few layers of tissue, and finally a piece of dampened blotting paper as the top layer. The cover is then put in place and sealed with masking tape. It is best to keep boxes of layered specimens in a refrigerator.

Reference: 424.

Some insects, such as small beetles, should be glued to triangles (see p. 28) directly from the layers for permanent preservation, but if they are to be pinned or otherwise treated, they must be relaxed as described on page 24.

Papering

Although pinning specimens when they are fresh is preferable, the storage method known as papering has long been used successfully for larger specimens of Lepidoptera, Trichoptera, Neuroptera, Odonata, and some other groups. It is a traditional way of storing unmounted butterflies and is satisfactory for some moths, although moths too often will have their relatively soft bodies flattened, legs or palpi broken, and the vestiture of the body partly rubbed off. To save space in most large collections, file Odonata permanently in clear plastic envelopes instead of pinning them.

Papering consists of placing specimens with the wings folded together dorsally (upper sides together) in folded triangles (fig. 15) or in small rectangular envelopes of glassine paper, which are the translucent envelopes familiar to stamp collectors. Glassine envelopes have become almost universally used in recent years because of the obvious advantages of transparency and ready availability. In many collections, glassine has become partly superseded by plastic. However, many collectors still prefer folded triangles of a softer, more absorbent paper, such as ordinary newsprint, and believe they are

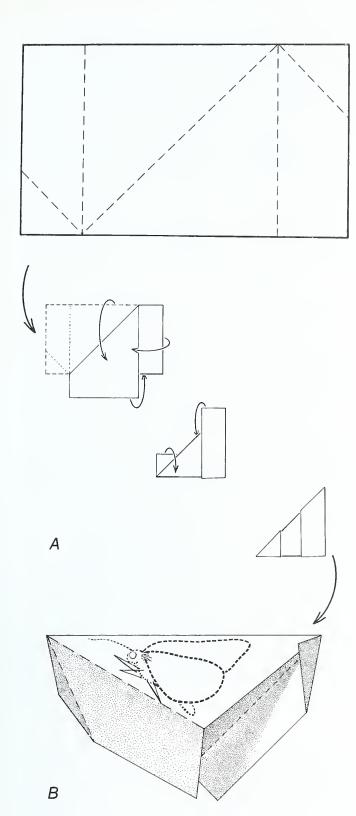


Figure 15. Method of folding a rectangular piece of paper to form a triangular envelope for temporary storage (papering) of large-winged specimens: *A*, Correct shape of unfolded paper, showing where folds should be made and sequence of first three folds; *B*, triangle almost completely folded, showing correct position of enclosed butterfly.

superior for preserving specimens. Specimens can become greasy after a time, and the oil is absorbed by paper such as newsprint but not by glassine. Moreover, glassine and plastic are very smooth, and specimens may slide about inside the envelopes during shipping, losing antennae and other brittle parts. Although softer kinds of paper do not retain creases well when folded, this shortcoming may be circumvented by preparing the triangles of such material well before they are needed and pressing them with a weight for a week or so. Triangles are easy to prepare if the paper is folded as shown in figure 15.

Some Lepidoptera are most easily papered if first placed in a relaxing box (see p. 24) for a day or two. The wings, often reversed in field-collected butterflies, may then be folded the proper way without difficulty. Do not pack specimens together tightly before they are dried or the bodies may be crushed. Do not store fresh specimens immediately in airtight containers or plastic envelopes or they will mold. Write collection data on the outside of the envelopes before inserting the insects.

Mounting Specimens

Specimens are mounted so that they may be handled and examined with the greatest convenience and with the least possible damage. Well-mounted specimens enhance the value of a collection; their value for research may depend to a great extent on how well they are prepared. Standardized methods have evolved over about 2 centuries in response both to the aesthetic sense of collectors and to the need for high quality research material. Although the style and technique of mounting may vary from one worker to another, the basic procedures outlined here are widely accepted. Methods of preparation are subject to improvement, but in the interest of uniformity it is best to follow currently accepted practices until the superiority of other methods has been proved.

The utility of a mounted specimen—that is, how well it is preserved, how safe it is from damage, and how much of the specimen can be examined conveniently—is generally of more importance than its beauty. Research considerations should take second priority only with specimens mounted specifically for non-technical display purposes.

Preparation of specimens for a permanent collection is discussed here except specimens to be kept permanently in a liquid preservative or in papers or envelopes (see Temporary Storage of Specimens, p. 22). Specimens to be prepared for a permanent collection may be fresh, that is, their body tissues not yet hardened or dried; or they may have been in temporary storage and must be specially treated before mounting. Dry specimens usually must be relaxed, and those preserved in liquid must be processed so that they will dry with minimal distortion or other damage.

Preparing Dry Specimens for Mounting

Any dry insect that is to be pinned must be relaxed, that is, remoistened enough to soften so that it will not break when the pin is inserted or so that parts of the specimen may be rearranged or repositioned. Insects, especially Lepidoptera, that are to have their wings spread should be relaxed even if they have been killed for only a short time. The muscles of Lepidoptera, once the stiffening of rigor mortis sets in, which occurs in a matter of minutes, are strong enough so that adjustment of the wings is difficult, but treatment in a relaxing chamber usually will make this procedure much easier. Eight hours in a relaxing chamber should suffice, but larger specimens may require 24 hours or more. Simply leaving specimens in a cyanide jar for awhile sometimes will relax them, but this method is not reliable.

Reference: 255.

High humidity must be provided in a relaxing chamber for periods varying from several hours up to about 3 days, depending on the circumstances, without the specimens actually becoming wet. The growth of mold is also to be avoided, since it will ruin specimens left too long in relaxing chambers unless a chemical mold inhibitor has been added. Insects killed with cyanide usually can be relaxed easily, but some killing agents, especially chloroform, ether, and carbon tetrachloride, may harden muscles to such an extent that the specimens are brittle and seemingly impervious to the humidity of the relaxing chamber. In Korea, for example, butterflies are injected in the thoracic muscles with very hot water through a fine hypodermic needle before spreading. Occasionally, however, some specimens can not be relaxed satisfactorily by any method.

Many kinds of receptacles can be used as relaxing chambers, including glass dishes or jars with covers (low, widemouthed jars or casserole dishes are excellent), tobacco or biscuit tins, even earthenware crocks. Glass or earthenware containers are not so immediately affected by fluctuations in temperature as are other types and thus may relax insects more evenly. Containers 5-15 cm deep are most convenient; clear plastic sandwich boxes not more that 2.5 cm deep will serve for small specimens if they are not on pins. A layer of damp sand, peat, or crumpled paper toweling is placed in the bottom of the container and covered with a layer of cotton, cellulose wadding, or jeweler's cotton. This layer will not absorb water readily and will prevent direct contact between the insects and the moisture beneath.

Some workers object to the use of cotton because of the tendency for insect legs to become entangled in it and break off. If this is a problem, cover the cotton with a single piece of soft tissue. For very small specimens, a lining of tissue or some absorbent material with a smooth surface is advantageous. Heavy paper such as blotting paper or cardboard may be used in place of cotton, but this should be supported 1 cm or more above the moist bottom layer to avoid direct contact. Wooden or plastic strips or fine-mesh plastic screen also may be used for this purpose.

Mold probably will not be a problem if insects are relaxed for no longer than 2-days at normal room temperature, but relaxing chambers in regular use should be kept clean, with frequent renewal of the contents. If mold is likely to develop, as may happen with large specimens held more that 2 days, a few crystals of naphthalene, paradichlorobenzene, phenol, or chlorocresol may be sprinkled in the bottom of the relaxing chamber, or a little thymol, which is more potent, may be used. All these chemicals, however, may damage plastic boxes.

Insects held too long in a killing jar, or those that were originally papered, pinned but unspread, or layered (that is, placed in small boxes between pieces of soft tissue) may be relaxed by placing them in a relaxing chamber. Papered specimens will relax faster if removed from their envelopes. For beetles and other insects that do not need to have the wings spread, holding them overnight or at most for 24 hours in a relaxing chamber will suffice. Small moths and delicate Neuroptera also should be relaxed sufficiently after 12-24 hours to allow the wings to be spread. Large moths, however, may take 48 hours or longer if the relaxing chamber is kept at room temperature. The process can be hastened and the chance of mold developing greatly reduced if the relaxing jar is subjected to a slight raising and lowering of temperature, as perhaps between 18° and 27° C. The process is greatly accelerated if the relaxing jar is set in, or floated on, warm water for an hour or more; specimens may be relaxed within 3-6 hours in this way. If the warm-water treatment is overdone, the specimens may be spoiled by the absorption of too much moisture. Some colors, especially nonmetallic greens in Lepidoptera, are unstable and may be completely bleached by exposure to too much humidity. Such material requires special attention; the specimens should be left in the relaxing chamber for the shortest possible time and ideally should be pinned and spread when fresh. Experience soon enables one to judge the best procedure for the particular kind of material being prepared.

The length of time that insects may be left safely in a relaxing chamber depends somewhat on the temperature. At 18°-24° C, they may be left for about 3 days, but beyond that time, they will begin to decompose. If the relaxing chamber is placed in a refrigerator at 3°-4°, the specimens may be kept for 2 weeks, although they may be slightly damaged from excessive condensation by that time. If relaxing chambers containing fresh specimens are placed in a deep freeze at -18° or lower, the specimens will remain in comparatively fresh condition for months, but not indefinitely. Specimens gradually desiccate and eventually will become dried. However, a freezer may be used to keep them fresh for a month or two and is a great convenience.

Even when specimens have been relaxed suitably for spreading, the wings may still seem stiff. In this instance, the wing muscles must be loosened by forcing the wings to move up and down. This may be safely done by pressing the tips of curved forceps firmly against the costal vein very near the base of the wing. The forceps should have the tips ground or honed smooth and not too sharp. Repeat this procedure separately with all four wings or they will revert gradually toward their original positions. With care, all the wings may be loosened in this way without leaving any visible marks.

Occasionally it may be desirable to relax and reposition only a part of an insect as, for example, moving a leg that may be concealing characters needed for identification. This may be accomplished by putting a drop or two of Barber's fluid (see Appendix) or ordinary household ammonia directly on the leg. Most household ammonia is now furnished with a detergent, which helps it wet and penetrate insect tissue. After a few moments, perhaps after adding a little more fluid, the part may be pried carefully with a pin. When it moves easily, it may be placed in the desired position, held there with a pin fixed in the same substrate as is holding the pin on which the specimen is mounted, and left until the fluid dries thoroughly.

A few methods of relaxing insects with heat have been used. These and others are summarized in the following reference, in which a steam-bath method is described.

Reference: 442.

Preparing Liquid-Preserved Specimens

Most specimens preserved in fluid must be removed from the liquid in which they have been stored so that they will dry with as little distortion or matting of hairs as possible. Only specimens with hard exoskeletons, such as beetles and some bugs (Pentatomidae, Cydnidae), may be mounted without special treatment when removed directly from the preserving fluid onto pins or points. The following methods have been used routinely for removing specimens from the usual fluid preservatives, and the specimens are often left in better condition than if they had been pinned while fresh, especially small Diptera.

The following equipment is needed: (1) A few screw-top jars about 5 cm in diameter with a cork cemented with epoxy on the top of each lid and a label on the outside showing clearly what they contain—some about one-third full of Cellosolve (2-ethoxyethanol, ethylene glycol ethyl ether) and some about one-third full of xylene; (2) a small dish, such as a watchglass; (3) absorbent tissue from which to twist small "pencils" for absorbing xylene; (4) insect pins or double mounts (see p. 28) for mounting the specimens; (5) adhesive in a jar with a rod in its stopper (see p. 28); (6) narrow-pointed forceps; and (7) a few small cards of blotting paper.

When specimens are ready for preparation, remove as many from the preservative as can be pinned or placed on triangles or card points in an hour (experience will tell). Place them on blotting paper, then drop them from the blotter into a jar of Cellosolve, and place a label with the collection data on the pin stuck into the cork cemented to the lid of the jar. This may be done at the end of the day and the specimens left in the Cellosolve overnight, or otherwise for about 3 hours, longer for large specimens. They may even be left over a weekend. The same jar of Cellosolve may be used several times. up to about 10 times if the insects are small. This part of the treatment removes water and other substances from the specimens. However, Cellosolve does not evaporate readily, so it must be removed subsequently with another solvent, which will evaporate readily.

The next step is to use forceps to remove the specimens carefully from the Cellosolve, place them again briefly on blotting paper, then into a jar containing xylene. The identifying label on the pin in the cork must also be transferred. Small specimens should be left in the xylene for about 1 hour, larger specimens for up to 4 hours. Specimens left too long in xylene will become extremely brittle and can hardly be put on a pin or triangle without losing legs, antennae, or the head. As with the Cellosolve, the xylene also may be used many times until it becomes so contaminated with Cellosolve that specimens dry slowly when removed from it. While specimens are still wet with Cellosolve or xylene, they are somewhat pliable, and legs and antennae may be repositioned slightly.

When specimens have been in the xylene for at least 1 hour, they may be mounted. Take the smallest ones first to avoid leaving them in the xylene too long. Remove them with small forceps and place them in a dish. The forceps will pick up a small amount of xylene, and the specimen will be left lying in it. While there, it may be positioned correctly for mounting; the wings will float out flat, sometimes with a little adjustment with a pin or the tip of the forceps. When it is positioned correctly, take a "pencil" of absorbent tissue and touch it to the specimen to remove the excess xylene. Larger specimens may be pinned directly in the usual manner (see p. 26). Just before the xylene fully dries from the surface of a small specimen, the tip of a triangle or a tiny pin called a minuten, already attached to its carrying insect pin, should be touched to adhesive (see Double Mounts, p. 28). The tip of the triangle may then be touched to the specimen, picking it up. If a minuten is used, it may be inserted into the thorax of the specimen. A little final adjustment of position may then be made, and the specimen is ready for its label and place in the collection. If the specimen has been placed on a minuten, having touched the tip of the minuten to the adhesive will leave a small amount of adhesive around the place where the minuten has pierced the specimen and will keep it from working loose when fully dried.

Specimens placed on regular pins should have a small amount of adhesive placed around the site where the pin protrudes from the lower side of the specimen. Specimens pinned after having been in fluid preservatives do not cling as firmly to the pin as do those pinned fresh.

This treatment will leave surface pile, hairs, and bristles in a loose, unmatted, natural condition. Small specimens that shrivel considerably after having been pinned fresh will usually dry in better condition if pinned or placed on triangles after this treatment.

Warning: Xylene is now considered to be carcinogenic. A new and already widely used chemical, Histo-Clear, is a promising substitute.

Reference: 372.

Direct Pinning

This section pertains entirely to insects because mites should never be mounted on pins. Direct pinning refers to the insertion of a standard insect pin directly through the body of an insect. Only insect pins should be used; ordinary straight pins are too short and thick and also have other disadvantages. Standard insect pins are 38 mm long and range in thickness from size 000 to 6 or 7. Heads are now commonly made of nylon. but they may be of a type called "upset," that is, an integral head is made by mechanically squeezing out the end of the pin, or a small piece of metal is pressed onto the pin. A well-made upset head is considered by some entomologists to be best; other kinds of heads sometimes come off, leaving a sharp point that easily can pierce a finger. Recently, however, pins have become available with nylon heads attached rather firmly. Pins of No. 2 diameter are most useful (0.46 mm in diameter). Most entomologists avoid the very slender pins of size 000 to 1, preferring to use double mounts (see p. 28), but now that soft polyethylene or plastic foam is commonly used for pinning bottoms in trays and boxes, these smaller sizes are not so impractical as formerly. Pins of larger diameter, Nos. 3-7, may be needed for large insects.

Standard insect pins are currently made of either ordinary spring steel, which is called 'black,' or stainless steel and with either a blued or a lacquered (japanned) finish. The black pins may corrode or rust with even slight exposure to moisture or to the body contents of the insects. Although the stainless steel pins are more expensive than black pins, their being rustproof makes them desirable for use in permanent collections. However, their points are somewhat more easily turned than those of black pins in piercing an insect with a hard cuticle, and they are not as rigid. For that reason, it is sometimes advisable to pierce an insect having an especially hard cuticle with a strong steel pin before inserting a stainless steel pin. Lacquered pins have a surface on which the insect may be less likely to become loose than it might on a bare pin.

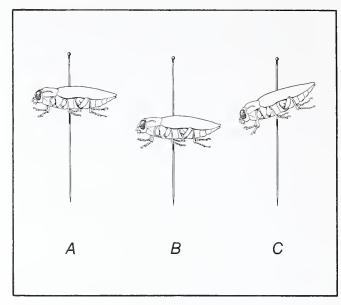


Figure 16. Right and wrong methods of pinning insects: *A*, Correct height and position; *B*, too low on pin; *C*, tilted on pin.

Insect pins made of German silver or brass were once common. They quickly corroded from the action of the insect body contents, producing a greenish verdigris about the pin in the insect and eventually eating entirely through the pin.

One who handles a large number of pinned specimens may find pinning or dental forceps helpful. Their curved tips permit the pin to be grasped below the data labels and enable one to set the pin firmly into the pinning-bottom material without bending the pin. The forceps are also of much assistance in removing pins tightly corroded into the cork pinning bottoms. The pin is grasped tightly above the cork and turned a little before it is lifted. However, with wings of most Lepidoptera, it is impractical to place pinning forceps below the specimen.

Insects should be pinned vertically through the body with a pin of appropriate thickness, using care that the pin does not tear off any legs as it goes through the body. Most insects are pinned to the right of the midline so that all the characters of at least one side will be visible. Figure 16 illustrates some right and wrong examples of pinning. Do not attempt to pin specimens unless they are relaxed (see p. 24) or freshly killed. Inserting a pin into a dry specimen may cause it to shatter. When pinning relaxed specimens or specimens taken from Cellosolve and xylene, a little glue may be needed where the pin emerges from the specimen to prevent the specimen when dry from working loose and rotating on the pin. Application of adhesive is unnecessary when mounting freshly killed insects.

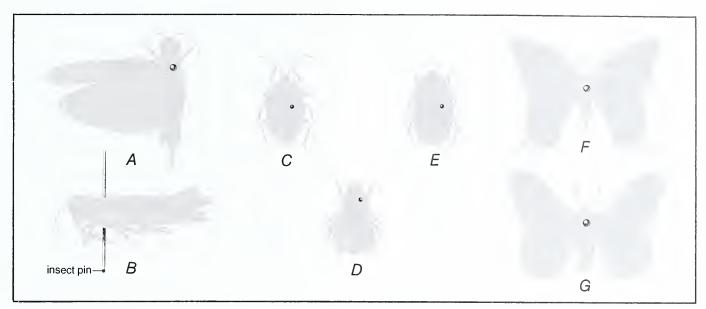


Figure 17. Examples of correct pinning methods for common insects; black spots show where pins should go. *A*, Grasshopper and related Orthoptera, showing how wings should be spread; *B*, side view of grasshopper, showing position of legs and antennae; *C*, stinkbug, order Heteroptera, showing method of pinning large

bugs; *D*, bee, order Hymenoptera, showing where bees, wasps, and flies should be pinned; *E*, may beetle, order Coleoptera, showing method of pinning beetles; *F* and *G*, butterfly and moth, order Lepidoptera, showing position of wings and antennae.

Standard methods of pinning some of the commoner types of insects are as follows:

- (1) Orthoptera—Pin through back of thorax to right of midline (fig. 17, *A-B*). For display purposes, one pair of wings may be spread as shown, but many orthopterists prefer to leave wings folded because of limited space in most large collections (see ref. 27).
- (2) Large Heteroptera—Pin through triangular scutellum to right of midline (fig. 17, C). Do not spread wings.
- (3) Large Hymenoptera and Diptera—Pin through thorax between or a little behind base of forewings and to right of midline (fig. 17, *D*). So that no characters on body are obscured, legs should be pushed down and away from thorax, and wings turned upward or sidewise from body. Wings of most Diptera will flip upward if specimen is laid on its back before pinning and pressure is applied simultaneously to base of each wing with pair of blunt forceps. Wings should be straightened if possible so venation is clearly visible. Folded or crumpled wings sometimes can be straightened by gentle brushing with a camel's hair brush dipped in 70 percent alcohol. For Hymenoptera wings, Peterson's XA mixture (xylene and ethanol, equal parts by volume) is recommended.
- (4) Large Coleoptera—Pin through right wing cover near base (fig. 17, E). Do not spread wings.
- (5) Large Lepidoptera and Odonata—Pin through middle of thorax at thickest point (fig. 17, F) or just behind

base of forewings (fig. 17, G). Spread wings as described on page 32.

The height of the insect on the pin will depend somewhat on its size, but enough of the pin should always be exposed above it to be grasped without the fingers touching and possibly damaging the specimen. Those mounted too high on a pin very likely will be damaged in handling. If pinned too low, the legs may be broken when the pin is inserted in a tray or box and insufficient space may be left for labels.

After the pin is inserted and before the specimen is dry, the legs, wings, and antennae should be arranged so that all parts are visible for study. With most insects, it is necessary only to arrange the legs and antennae in the desired position and let them dry, but occasionally it is necessary to hold the appendages in place with insect pins until the specimen is dry. With long-legged species or those with drooping abdomens, the legs and abdomens may be supported until dry with a piece of stiff paper pushed up on the pin from beneath. Once the specimens are dry, this paper support can be removed. For moths, butterflies, and other insects that should be mounted with the wings spread, use a spreading board (see p. 30) or spreading block (see p. 30).

Although some entomologists glue small insects directly to the side of a standard insect pin, this practice is not recommended because too much of the insect is often obscured either by the glue or by the pin, and the adhesive does not adhere well to the pin. For small insects, use a double mount.

Double Mounts

Insects that are too small to be pinned directly on standard pins and yet should be preserved dry may be pinned as double mounts. This term refers to the insect's being mounted on a minuten or card point, which in turn is mounted or attached to a standard insect pin (fig. 18, A). Minutens are available from supply houses in 10and 15-mm lengths and in two or three thicknesses. They are finely pointed at one end, headless on the other, and generally of stainless steel. Double mounts are assembled by inserting the minuten into a small cube of soft, pithy material such as fine cork, balsa wood, fine-textured plastic, or polyporus, which is a pure white material obtained from a bracket fungus. Polyporus traditionally has been a favorite material, but it is expensive and difficult to obtain, especially in America. Many entomologists prefer silicone rubber, obtained from plastics suppliers and made into plaques by pouring the polymerized material, a thick creamy liguid, into a flat-bottomed plastic container to a depth of about 2.5 mm and allowing it to solidify for several hours. It may then be lifted easily from the mold and cut with a sharp knife or razor blade into square strips and finally into cubes. With most materials, the minuten must be inserted point first, but with silicone rubber it may be inserted dull end first until it strikes the surface on which the cube is lying, and it will be held firmly. Minutens should be handled with forceps; they are so small that even the unsharpened end can easily pierce a finger.

It is possible, and sometimes preferable, to mount an insect on a minuten before inserting the minuten into the mounting cube; however, it is most convenient to prepare a series of minuten mounts beforehand, already attached to standard No. 3 pins. To mount extremely small insects, such as tiny parasitic wasps, on minutens, pick up a droplet of cement with the prepared minuten and simply place the tip of the minuten with the cement on it between the base of the insect legs. In mounting an insect on a minuten, the pin need extend no more than barely through the insect. If the insect is lying on a glass surface when it is pierced with the minuten, a little extra pressure will curl the point of the minuten back into the insect and insure that the specimen will not come off the minuten.

Many entomologists prefer to mount insects on a minuten in a vertical position in a short strip of polyporus or silicone, with the minuten therefore parallel to the main pin. The insect lies sidewise in the finished mount, in an excellent position for examination under a microscope, and is less liable to damage in handling than it would be otherwise.

Reference: 346.

Card points are slender little triangles of stiff paper. They are pinned through the broad end with a No. 2 or 3 insect pin, and the insect is then glued to the point (fig. 18, *B*). Card points may be cut with scissors from a strip of paper; they should be no more than 12 mm long

and 3 mm wide. However, a special punch for card points, obtainable from entomological supply houses, will make better, more uniform points. Card points should be made only from good quality paper, as good as or better than that used for data labels (see p. 40). If specimens are in good condition and are well prepared, they may reasonably be kept in museum collections for a long time, perhaps even for centuries. Much of the paper in common use does not have that kind of life expectancy; it becomes yellow and brittle with age. Paper made especially to last, such as that used for herbarium sheets in botanical collections, is highly recommended.

For similar reasons, the choice of the best adhesive for card points may be equally important, but unfortunately the aging properties of various glues are not known. Ordinary white (casein) glue, clear acetate cement, or fingernail polish is used commonly; white shellac is less satisfactory because it is usually too thin and flows over a specimen when applied.

Entomologists of the USDA Systematic Entomology Laboratory have for many years used a very viscous polyvinyl acetate for double mounting and similar uses. It is obtainable in granular, bead, or pellet form. A small quantity is placed in a bottle with a glass rod in its stopper and covered with absolute ethanol. It will dissolve in a day or two into a thick solution. If it is too thick, it will "string out," and more ethanol should be added. If it is too thin, the bottle should be left open to allow some of the ethanol to evaporate. After a period of use, the solution will also normally become too thick, and then more ethanol must be added. Specimens adhere very well to a pin or a point with this solution, and they may be removed with 95 percent ethanol.

Many entomologists use shellac gel (see ref. 285). Because its preparation involves boiling white shellac solution, a procedure with some danger from fire, it is not recommended here. However, a safer method of preparation may eventually be found for this useful medium.

Whatever adhesive is used, it should not be permitted to get so thick that it "strings." Should this happen, add a little solvent to the adhesive until it attains the proper consistency. Nor should it be so thin that it flows over a specimen. Only a small amount of adhesive should be used to glue the specimen to the card point, since excessive glue may obscure certain sutures or sclerites necessary for identification, just as the card point may conceal certain ventral structures if allowed to extend beyond the midline of the insect.

For most insects, the card point is attached to the *right* side of the specimen (fig. 18, *C*), with the left side and midventral area clear. For better adhesion with some insects, the tip of the card point may be bent downward at a slight angle to fit against the side of the specimen. Only a very small part of the point should be bent. With a little practice it will be easy to judge how much of the point to bend and at what angle to fit the particular insect being mounted.

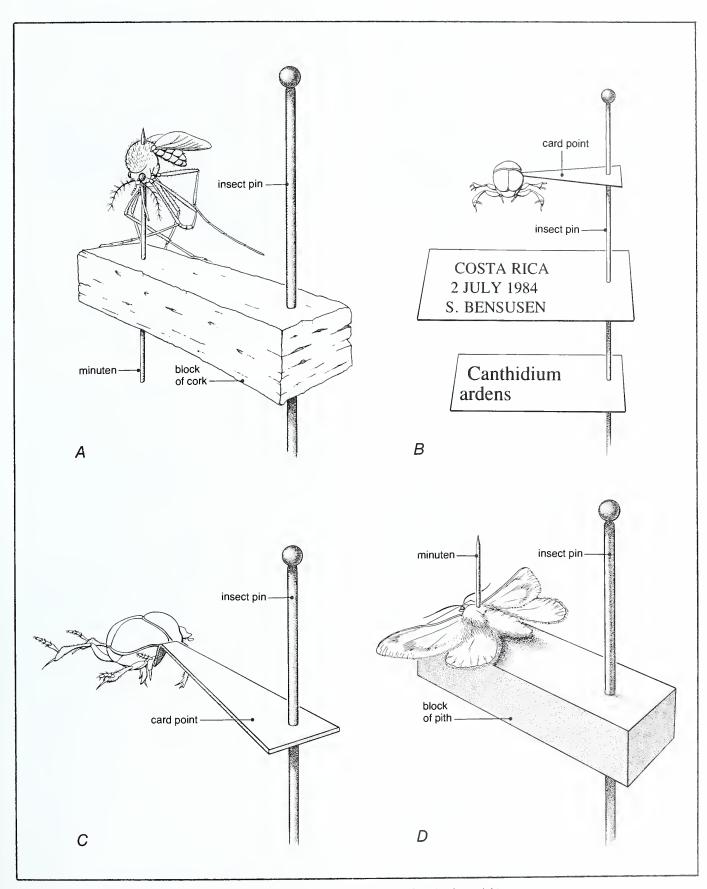


Figure 18. Double mounts for small insects: *A*, Mosquito pinned with minuten to block of cork on regular insect pin; *B*, position of card point and

labels on pin; *C*, attachment of card point to right side of specimen; *D*, small moth pinned with minuten to block of pith on regular insect pin.

One method to insure that the specimen is oriented properly on the point is to place it on its back with its head toward you; then with the pin held upside down, touch a bit of adhesive to the bent point and apply it to the right side of the insect. If the top of the point can be slipped between the body of the insect and an adjacent leg, a stronger mount will result. The card point should be attached to the side of the thorax, not to the wing, abdomen, or head. Some insects, such as small flies and wasps, are mounted on unbent points. Those working with small flies prefer to attach the card point to the left side of the specimen with the legs facing the pin.

Opinions differ on when to use direct pinning and when to use a double mount, and perhaps this is best determined through experience. A general rule of thumb is that if you can mount a small insect on a size 1 or 0 pin without damaging the specimen, do not use a double mount. Insects too heavy to be held on the point by adhesive yet too small to be pinned with standard pins may be attached to card points by puncturing the right side of the insect at the place where the card point normally would be placed and inserting into this puncture the tip of an unbent card point with a little glue on it. For puncturing specimens, use a needle ground and polished to make a small, sharp scalpel. Some specimens, such as moths, should never be glued to points; other specimens should never be pinned with minutens. The following suggestions will serve as a guide:

- (1) Small moths, caddisflies, and neuropteroids— Mount on minuten inserted through center of thorax with abdomen positioned toward insect pin (fig. 18, *D*). Mount must be sufficiently low so that head of pin can be grasped easily with fingers or pinning forceps. Do not glue small moths to points. Ideally, such specimens should be spread in the conventional manner despite their small size.
- (2) Mosquitoes and small flies (freshly killed)—Pin with minuten through the thorax with left side of specimen positioned toward main pin (fig. 18, A). Note that minuten is vertical, which is more advantageous than if it were horizontal because specimen is less liable to come into contact with fingers or pinning forceps. Placing a small amount of glue on tip of minuten before piercing specimen will help hold soft-bodied insects.
- (3) Small wasps and flies (not freshly killed)—Mount on unbent card point with point inserted between coxae on right side of insect, keeping clear of midline, or glue tip of point to mesopleuron.
- (4) Small beetles, bugs, leafhoppers, and most other small insects—Glue card point with tip bent down to right side of specimen (fig. 18, *C*).

As to the length of pin exposed above the specimen, double mounts should conform to the same rule as in direct pinning: Do not place a double mount too high on the pin. It must be possible to grasp the head of the

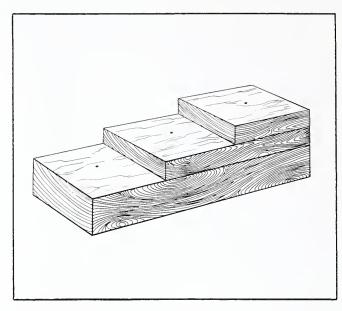


Figure 19. Pinning block for adjusting specimens and labels to uniform height on pin.

pin between the thumb and index finger without touching the specimen. Uniform height may be obtained by using a simple measuring device such as a three-step block (fig. 19). Double-mount cubes or points may be adjusted at any time, whereas once a directly pinned insect has dried on a pin, it is virtually impossible to move it without damage. If points become loose on the main pin, place a little adhesive at the connection.

Reference: 45.

Spreading Boards and Blocks

All insects preserved with the wings spread uniformly are set and dried in this position on spreading boards or blocks; spreading boards are more commonly used than spreading blocks. Although such pinning aids vary greatly in design, the same basic principle is inherent in all, that is, a smooth surface on which the wings are spread and positioned horizontally; a central, longitudinal groove for the body of the insect; and a layer of soft material into which the pin bearing the insect is inserted to hold the specimen at the proper height. An active collector will need from several to many spreading boards because the insects must dry for a considerable time (about 2 weeks for most specimens, even very small ones) before being removed from the boards. Spreading boards may be purchased from biological supply houses or may easily be made as described here if the proper materials can be obtained. When purchasing spreading boards, avoid (1) too hard or too soft a material for the pinning medium under the central groove, (2) too hard an upper pinning surface, and (3) top pieces without the same thickness at the center (an especially common fault in beveled boards). This last defect may be corrected by sanding down the higher side; evenness is especially critical when working with small specimens.

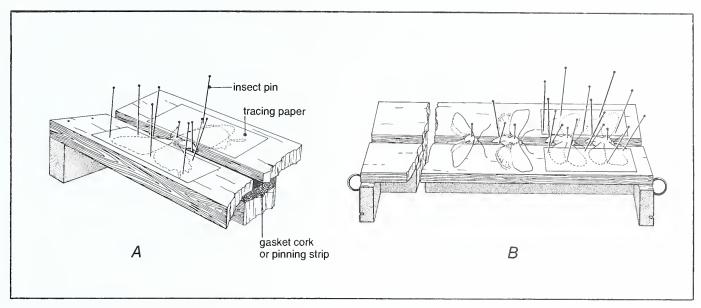


Figure 20. Spreading boards, showing (*A*) construction and how butterfly wings are spread and abdomen and antennae are arranged; and

(B) how several specimens of about same size can be prepared simultaneously.

Construction of Spreading Boards. A spreading board of simple design (fig. 20, A) requires the following materials:

(A) Two top pieces, 9 mm by 4.8 cm by 38 cm, preferably of seasoned basswood, a fine-grained, durable wood from trees of the genus Tilia. Holes made in it by insect pins tend to close after they are removed. If the surface of the board is lightly sanded after use, especially when working with small specimens, its smooth, even quality can be maintained through many years of use. Basswood is sometimes known as 'whitewood'; however, wood from trees of the genus Liriodendron is also sold under this name. If basswood cannot be obtained, well-seasoned white pine selected for softness and 'pinnability' is serviceable. A third choice is 20-cm (8-inch) beveled redwood siding. Beveled top pieces are desirable because the beveling, sloping inward, compensates for the tendency of the wings of spread specimens to droop slightly after the specimens are removed from the board. The 20-cm siding is actually 19.1 cm wide, and a 0.9-meter piece of it will provide two pairs of top pieces. One pair cut slightly more than 4.8 cm wide from the wide side of the board and planed to exact width will make a pair 11 mm thick at the narrow side, and another pair cut from the same side of the board will provide a second pair about 8 mm thick at the narrow side. Redwood is stiff and fine grained, but it splits and splinters easily.

- (B) Two end pieces of any good, fine-grained wood, 2 cm square by 10 cm.
- (C) One strip of entomological or gasket cork or foam, 6 mm by 3 cm by 34 cm.

(D) One base of plywood or any fine-grained wood, 6 mm by 10 cm by 38 cm.

These materials are for a spreading board with a central groove 6 mm wide. Boards with grooves of several sizes will be needed. For the larger Lepidoptera (macrolepidoptera or 'macros'), the most useful widths are 3, 6, and 9 mm. For very large moths, a width of 17 mm is required; the board will also have to be as much as 15 cm in total width with a groove depth of 16 mm. For small moths (microlepidoptera or 'micros'), special boards with groove widths of 1.5-2 mm will be needed, with the groove shallow enough for minutens, and the width and thickness of the top pieces (A) must be altered accordingly. The pinning medium (C) could be of polyethylene foam, but to give specimens firm support, the entire depth between the top pieces and the base would have to be filled with the material. A dense, finely textured plastic foam known as 'Plastazote' is better than polyethylene for entomological applications and is available in Britain but so far not in the United States. A strip of modelmaker's balsa wood, selected for pinning softness, may also be satisfactory.

The end pieces (B) should be glued with epoxy or other good adhesive and nailed to the top pieces (A) with the proper groove width maintained. Then the pinning strip (C) should be firmly glued to the underside of the top pieces (A), the same side on which the end pieces (B) were fastened, and should cover the central groove. Finally, after the adhesive has set. the base (D) should be attached. If it is affixed to the end pieces (B) with two flat-headed wood screws (about No. 5, 19 mm) countersunk into the base piece and screwed into each end piece, the base may be removed easily later if replacement of the pinning strip is necessary.

Using the Spreading Boards. Before spreading specimens, the spreading boards and the following materials should be at hand:

- (1) Pins (called setting pins) of size 00 or 000 for bringing wings into position. Setting pins used by some lepidopterists are made by inserting a minuten into a round matchstick and securing it with a drop of glue.
- (2) Strips of glassine or tracing paper (the translucent, smooth paper used for tracing, not what a draftsman calls tracing paper). Cellophane, plastic film, or waxed paper should not be used. Their disadvantages include expanding with moisture and becoming electrostatically charged or containing a substance that pulls scales off the wings. The strips of tracing paper should be wide enough to extend from the base to a little beyond the end of the wings of the specimens being spread. Strips about 25 mm wide are convenient for spreading most Lepidoptera. Short ones are used when spreading specimens that have been relaxed from a dried condition, but strips long enough to cover several specimens in a row on the board are commonly used for freshly caught insects. The strips are often used with a narrow fold alongside the body of the specimen with the fold upward; this provides a rounded edge that reduces the likelihood of a sharp edge displacing a row of scales. This fold may be made by holding the strip on a spreading board with 3-5 mm of it overhanging the edge of board, running a finger along the overhang to bend it down, and then firmly folding it back.
- (3) Glass-headed pins at least 2.5 cm long for holding the strips in place. Ordinary No. 2 or 3 insect pins with nylon heads may also be used, but some lepidopterists find them hard on the fingers.

With this equipment ready, the collector is prepared to mount and spread the specimens (fig. 20, B). The specimens must be properly relaxed (see p. 24), even the freshly collected ones, before any attempt is made to spread the wings. Insert an insect pin of appropriate size through the middle of the thorax, leaving at least 7 mm of pin above the specimen. The pin should pass through the body as nearly vertically as possible to avoid having the wings higher on one side than on the other. Pin the specimen into the central groove of the spreading board so that the wings are exactly level with the surface of the board. Carefully draw each wing forward with the point of a setting pin inserted near the base and immediately behind the strong veins that lie near the front of the wings. If care is taken not to tear the wing, the fine setting pins should leave holes so small that they are barely visible. The hindmargin of the forewings should be at right angles to the groove in the board. Bring the hindwing into a natural position with its base slightly under the forewing. The setting pins will hold the wings in position until they can be secured with the paper strips.

The strip is placed close to the body of the insect, with its fold upward and toward the insect. A glass-headed pin is inserted in the middle of the folded part of the strip just outside the margin of the forewing. The pin may be tilted slightly away from the wing to keep the strip down against the wing. The strip is then carefully stretched backward and another pin placed just behind the hindwing. A third pin in the notch where the forewings and hindwings meet is usually enough. None of the three pins on each side of the specimen should pass through the wings. Once the paper strips are in place, the setting pins may be removed. Twisting the setting pins a little as they are removed will prevent a possible bent tip from hocking onto a wing vein and pulling the whole wing out of place.

Fresh specimens may be arranged closely on the board in series of five, six, or even more before the paper strips are applied to cover all. Relaxed specimens, however, should be treated individually because they dry so quickly that antennae may break or the wings curl if the spreading is not completed promptly.

To prevent the abdomen from drooping as the specimen dries, support it with a pin on each side, crossing beneath. Pins may also be used to arrange and hold the antennae and legs in position until they dry. The appearance of many insects may be improved by gently blowing on them before spreading to remove extraneous loose scales and to straighten the hairs or, with small moths, the fringes of the wings. In working with small insects, a large magnifying lens mounted on an adjustable stand may be very helpful.

Specimens relaxed from a dried condition present some additional problems. The wings may be stiff and require loosening (see p. 25). If the wings of a relaxed specimen are turned upward and do not lie on the surface of the spreading board, the paper strips may need to be pinned over the wings to hold them down before they are positioned. Since the wings of relaxed specimens are still relatively stiff, skillful manipulation is needed to spread the wings without tearing or leaving excessively large pinholes. If the wings do not move readily under gentle pressure, do not force and possibly break them. Return the specimen to the relaxing chamber.

Construction of Spreading Blocks. The spreading block is a modification of the spreading board designed to accommodate only one specimen at a time. Blocks often are preferred by specialists in microlepidoptera but can be used for other insects as well. The design is simple (fig. 21), consisting of a wooden cube about 3 cm on a side for most insects, with a groove across the middle of one face. The width and depth of the groove vary to suit the size of the insect to be spread, usually 1.5-2 mm in width and deep enough to accommodate a strip of fine cork, polyporus, or similar pithy material into which the pin or minuten is lodged to hold the insects being spread. The groove should be cut parallel with the grain of the wood, and the top surface of the block should be sanded exceedingly smooth. Before

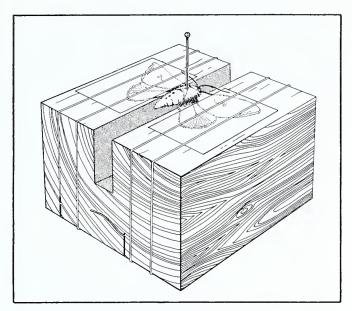


Figure 21. Construction and use of spreading block for small insects.

the pinning strip is wedged or cemented into the bottom of the groove, a hole about 1 mm in diameter should be drilled squarely in its middle. The pin can extend into this hole when the insect becomes level with the spreading surface. A few gashes, made by pressing the blade of a thin knife in the upper corners of the block near each end of the groove, should be made to catch the thread that will hold the wings of the insect.

The insect to be spread, mounted either on a standard insect pin or on a minuten, is pinned into the groove as with a spreading board, and the wings are manipulated by gentle blowing and using a setting pin. A piece of fine silk or nylon thread is then caught in one of the knife gashes and brought over the wings, and, if necessary, once around the block and again over the wing at another point, and finally caught again in the knife gash. A small piece of tracing paper may be placed on the wings before passing the thread over them, but if special scale tufts are found on the wings it is better to omit the paper and leave the tufts in a natural position.

Specimens either on spreading boards or blocks should be placed in a warm, well-ventilated place to dry for at least 2 weeks. Even very small moths require that long to dry completely and become stiff. If they are placed in a low-temperature oven, such as is used for drying plant specimens, 2 days may suffice. Specimens relaxed from a dry condition, as already noted, dry quickest, but even they should be left for several days. Fresh specimens, even large ones, may be dried in 2 days or less with heat. Where humidity is low and there is ample sunshine, the spreading boards or blocks may be placed in cardboard cartons painted black and left out in full sunlight for about 2 days. Occasionally, spec-

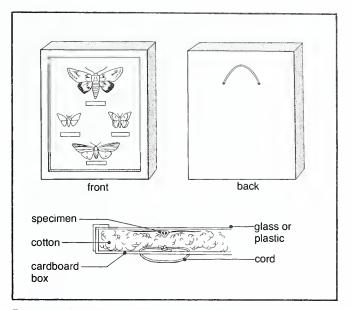


Figure 22. Riker mount for exhibiting specimens so they can be handled freely for close examination without risk of damage.

imens may become greasy, but otherwise no harm results. The spreading boards or blocks must be kept where they are safe from mice, bats, dermestid beetles, lizards, psocids (booklice and barklice), and ants, especially in the Tropics. One preventative measure that is sometimes advisable is to place the boards or blocks on bricks set in pans of water. If they are hung from the ceiling, a mosquito net around them may be necessary.

Always keep temporary data labels with specimens on spreading boards or associated with them in some way to insure that there is no confusion or loss of data when they are removed from the boards.

Spreading is a highly individualistic skill, subject to wide variation. Nearly everyone, with practice, evolves his or her own technique, so that two workers may appear to follow different procedures and yet produce equally good results. There is no single standardized technique with respect to the fine points of spreading.

References: 264, 411, 412.

Riker Mounts

It is sometimes desirable to prepare specimens for exhibition in such a way that they may be handled freely for close examination without risk of damage. Riker mounts have long been used for this purpose. They may be purchased from entomological supply houses, but similar cases may easily be constructed. The Riker mount (fig. 22) is simply a flat cardboard box about 3 cm deep, filled with cotton, and having a pane of glass or plastic set into the cover. Unpinned specimens are

placed upsidedown on the glass of the cover, spread into position with some cotton held in place by small weights, and allowed to dry thoroughly (about 2 weeks). Then the weights are removed, enough cotton is added to hold the specimens firmly in place, a little fumigant is added to kill any pests or their eggs that might have been laid in the box, and the bottom part of the box is put in place. When the box is closed, it should be sealed completely to prevent access to pests. Plant material may also be dried in place with the specimens.

Riker mounts are practical only for relatively large insects, such as butterflies, larger moths, beetles, and dragonflies, that are suitable for such display. Although Riker-mounted specimens are useful for classroom instruction and general display, they are not used for storage of insects in a scientific collection, where specimens must be available for examination from all angles under magnification. Riker mounts should be inspected periodically for pests and kept out of sunlight, which will cause fading of colors and general deterioration.

It is sometimes desirable to put pinned specimens into Riker mounts. To do so, remove the pin or cut it off flush with the surface of the insect (see ref. 202).

Inflation of Larvae

A common practice in the 19th and early 20th centuries was to preserve larvae, mainly caterpillars, by inflation. That practice has largely been abandoned in favor of alcoholic preservation or freeze-drying. These latter methods permit more thorough examination of all parts of the specimens, even internal organs, which must be removed before inflation. Some of the colors of the larger larvae are better preserved in inflated specimens than in alcohol, but color photography has made preservation of the larval colors less essential. However, the technique is still potentially useful and, if well done, is not to be discouraged. For instructions on how to inflate larvae, the following references may be consulted.

References: 15 (pp. 69-70), 177, 285.

Artificial Drying

Many kinds of soft-bodied insects and other arthropods may be preserved in a very lifelike manner and with no loss of color by freeze-drying. This process was developed fairly recently and is still somewhat experimental in its use with insects. It is, however, a great improvement over traditional methods of preservation in fluids or by inflation, especially with lepidopterous larvae, in which important characters of color and pattern are largely lost in all the traditional methods. The cost of freeze-drying equipment is unfortunately high, amounting to several hundred dollars.

Briefly, the procedure consists of killing the insect by first freezing it in a natural position and then dehydrating it under vacuum in a desiccator jar kept inside a freezer at -4° to -7° C. With a vacuum of 0.1 micrometer at -7°, a medium-sized caterpillar will lose about 90 percent of its moisture and about 75 percent of its weight in 48 hours. Its frozen condition prevents distortion while drying. The time required to complete drying is variable, at least a few days with small specimens and more than a week with larger ones. When dry, they can be brought up to room temperature and pressure and permanently stored in a collection. Like all well-dried insect specimens, they are rather brittle and must be handled carefully. Freeze-drying has also yielded excellent specimens of plant galls formed by insects.

An inexpensive method of freeze-drying (ref. 129) requires about 100 days to dry a medium-sized larva. The use of acetone is recommended before drying pinned specimens for better preservation of their colors, one of the features sought in artificial drying (ref. 39).

Critical-point drying (ref. 160) was developed to prepare specimens for study under the scanning electron microscope. Like freeze-drying, it is expensive and sophisticated, but it is designed for specimens in fluid. With this method, the specimens, although dried, are left somewhat pliable.

As techniques involved in freeze-drying and criticalpoint drying are refined, it may become feasible to keep excellently prepared dry specimens of immature insects in entomological collections alongside pinned adults.

References: 39, 106, 129, 160, 184, 215, 468.

Embedding

Preservation of various kinds of biological specimens in polymerized transparent plastics was popular in the 1940's and 1950's and is still of some interest. The process is rather complicated and laborious, but if carefully done it will yield useful preparations, especially for exhibits and teaching. Directions for embedding insect specimens may be found in the following references and in directions furnished by suppliers of the materials.

References: 124, 131, 197.

Mounting Specimens for Microscopic Examination

The small size of mites, thrips, whiteflies, aphids, scale insects, fleas, lice, and many other insects, as well as the necessity of clearly seeing minute details of larger insects, requires examination under a compound microscope at high magnification. Such specimens and parts of specimens must therefore be specially prepared and placed temporarily or permanently on microscope slides. If large and thick or complex structures that must be examined from several angles make slide mounting inadvisable, they may be examined in a liquid and preserved in microvials. Whichever course is adopted, their preliminary treatment is the same.

The techniques and materials used in preparing specimens for high-power microscopic examination vary considerably according to the kind of insect or mite as well as the researcher's preferences. The information given here will provide the reader with a basic concept of the principles involved in preparing specimens for such study. For more specific instructions, consult the following references or write to the Biosystematics and Beneficial Insects Institute (see p. 97). For reagents and media formulas mentioned here, see the Appendix, and for preparation procedures, refer to Sample Procedures, p. 37.

References (general): 12, 63, 134, 172, 175, 209, 279, 304, 363, 387, 452, 463.

Although information on slide preparation is broad and varies considerably according to the condition of the specimen and the mounting medium used, certain features are common to all processes. Cleaning, clearing, and maceration are nearly always necessary preliminaries. It is often desirable to dissect and critically examine specimens after the preliminary treatment and before mounting.

Clearing is the process of making the tissues of the specimen more transparent. It is often advisable to remove internal organs and muscles by using chemicals and to extend, manipulate, or dissect the specimens. This chemical removal of muscles and other soft tissues is known as maceration, although it is sometimes incorrectly called clearing. The agents used to macerate specimens usually also clean and clear them. Many mounting mediums also act as clearing agents to some extent.

Reference: 190.

Dehydration is usually a necessary preliminary to mounting, especially if the medium has a resin base. With some kinds of specimens, it is advisable to do this gradually or in steps to avoid distorting the specimens.

Staining is sometimes necessary with insect and mite specimens because their immersion in the mounting medium may make colorless and transparent tissues virtually invisible if the medium has a refractive index close to that of the tissues of the specimen (see ref. 400). Bleaching, usually accomplished with hydrogen peroxide, may also be required in very dark-colored specimens.

Washing is usually necessary at one or more stages in the process to remove and prevent excessive action by certain reagents used.

The final stage in preparing permanent mounts is thorough drying or hardening of the medium. This may be done in any clean environment or in an oven under gentle heat. The mounts should be carefully labeled either before drying or afterward. If more than one

mount is being made at a time, some recognition mark or code must be used on reagent containers and anything associated with the specimen so that the final mount may be correctly labeled.

The following procedures are given for mounting specimens to be examined microscopically:

(1) Maceration. Since only the sclerotic or chitinized parts of the insects are ordinarily needed in a preparation, the aim of maceration is to eliminate external secretions, foreign matter, some organs, muscles, and fat bodies without damage to chitinous parts. This is accomplished by immersing the specimen in a suitable agent, such as a sodium hydroxide (NaOH) solution, lactic acid, or lactophenol. These chemicals are strongly caustic and must be handled carefully to avoid damage to the skin and eyes. If any is inadvertently spattered on the skin, immediately wash it off with water.

Although textbooks specify potassium hydroxide (KOH) for maceration, this chemical must be used cautiously because specimens may be easily and quickly damaged or completely ruined in it. NaOH will perform as well as KOH or even better. Fine ducts lost with KOH remain even after lengthy treatment with NaOH, which will damage only teneral or newly emerged specimens. The amount of time a specimen is left in the macerating agent depends on the degree of sclerotization and the age and pigmentation of the specimen. For some relatively large, whole insects, the cuticle must be punctured with a fine needle to allow the agent to penetrate the body. Heating accelerates the action, but care must be taken to avoid damage by excessive action, especially if the specimen is at all teneral. Never heat the genitalia of microlepidoptera as it distorts the sclerotized parts. Immersion of the genitalia in cold 10-20 percent KOH solution overnight is recommended instead. Boiling for a minute in a 10 percent solution of NaOH (ordinary household lye) will clear most other small genitalia. NaOH supplied by chemical firms in pellet form is most convenient; three pellets in about 10 ml of water may be used for a day. The solution, however, is useless if left overnight. Even if it boils dry on a hotplate set a little above its boiling point, a specimen in it will seldom be damaged by NaOH, although it will be completely dissolved in KOH. Adding water to a specimen boiled dry in NaOH solution will usually restore it.

For directions on how to macerate insect genitalia, see p. 37.

- (2) Washing. For the removal of the caustic agent used to macerate the specimen, ordinary tapwater in a small dish, such as a small plastic bottle cover, will suffice. Distilled water is unnecessary. If the specimen is placed for at least a few minutes in plain water for manipulation, dissection, or examination, it then will be ready for further treatment. Adding a drop of acetic acid (white vinegar) will guarantee that no caustic remains.
- (3a) Staining. After clearing and washing, specimens may be stained if necessary, although if a phase-contrast microscope is available, staining, even with colorless specimens, is unnecessary. Several kinds of stains are available from biological supply houses. Acid fuchsin is generally used for aphids, lice, and scale insects. Thrips and fleas should never be stained; most acarologists do not stain mites if they are to be mounted in Hoyer's medium. An easily obtained stain for the exoskeleton of insects is made by dissolving a small amount of Mercurochrome crystals in water. Specimens may be immersed in the stain solution for 1 minute or more, depending on the degree of staining needed, and then briefly rinsed in plain water.

References: 71, 146.

- (3b) Bleaching. If specimens are too dark to reveal sufficient detail after maceration, they may be bleached in a mixture of one part strong ammonia solution to six parts hydrogen peroxide solution. The length of time the specimen is left in the ammonia-peroxide solution depends on the amount of bleaching needed.
- (4) Mounting. At this point, further treatment depends on what use is to be made of the preparation. It may be needed only temporarily in routine work and may be discarded after examination, or it may be desirable to keep the preparation permanently, either in glycerin in a microvial or in a mounting medium on a slide. If it is to be kept in a microvial, see Preparation and Storage of Genitalia (p. 37); if it is to be mounted on a slide, further treatment depends on the mounting medium used.
- (4a) Temporary Mounting. A temporary mount can be made with lactic acid or other medium on a 2.5- by 7.5-cm cavity slide. The specimen is placed near the edge of the cavity and wedged into position by manipulating a cover glass over the cavity and the specimen. A fine needle will help bring the specimen into the desired position before the cover glass is centered over the cavity. Once the specimen is in position and the cover glass centered, a commercial ringing compound, nail polish, or quick-drying cement is used to seal around the edge of the cover glass. Such slides may be kept for a year or more, but because they take up more space in a collection than permanent slides, the specimens eventually are usually placed in vials of alcohol for storage.

Temporary mounts are advantageous in that the specimen can be turned easily and viewed from many angles. However, because of the thickness of the mounts, a vertical illuminator operated through a microscope or some alternate method of direct lighting is generally needed.

Genitalia and other insect or mite parts may be examined and drawings made with the aid of an ocular grid in the microscope while they are lying in water in the dish in which they were dissected and extended. Water gives contrast to the structure, which may be difficult to see in glycerin. The water should be "dead," that is, boiled to drive out gases that may form bubbles in or on the object. The object may be held in place with a minuten bent L-wise and laid over the object or by piercing it at a convenient place.

(4b) Mounting Media. The old standard medium for permanence is Canada balsam. If it is used, the specimen must first be dehydrated through a series of alcohols of increasing concentration. Furthermore, balsam yellows with age and makes photography difficult; it is also difficult to manipulate delicate specimens in it. The mounting medium should be selected after consulting with a specialist or by referring to textbooks. Mites, for example, require special treatment, mainly because their cuticle differs from that of insects.

The most satisfactory mounting medium for most insects (other than scales and thrips) is Euparal, a synthetic preparation used for many decades. When it was unobtainable, especially during the World Wars, an inferior compound was used. Euparal may be obtained from medical or entomological supply houses and other sources, all of which import it from Germany. Its formula is a proprietary secret. It is not necessary to dehydrate specimens before mounting them in it. Good preparations may be made from specimens taken directly from 80 percent ethanol and from specimens immersed in 95 percent ethanol for only a minute.

The medium is water-white, remains so indefinitely, and for remounting, in case of breakage, specimens may be removed by soaking in absolute ethanol. Euparal has a very decided advantage over other media in that small air bubbles trapped in slide preparations are absorbed by the medium during drying, although this sometimes requires several days. Its only disadvantage is that it shrinks considerably in drying. In moderately thick preparations, this results in shrinkage away from the edges of the cover glass. This may be countered by adding additional Euparal until there is no further shrinkage, or in many instances by using a large cover glass, 2.2 cm in diameter, which in drying will pull down around the edges instead of allowing the medium to draw inward. The medium is relatively fast-drying. Allowing the slide to remain overnight in an oven set at about 35° C or in the open at room temperature for a few days will yield usable and permanent preparations.

Hoyer's medium and polyvinyl alcohol (PVA) are aqueous mounting media. Slides made with them are considered only semipermanent, although in the U.S. National Collection at the Smithsonian Institution, some 40-year-old slides of mites mounted in Hoyer's medium are still in good condition. Slide preparations made with Hoyer's or PVA, particularly of large or thick specimens, tend to crystallize with age and may need remounting. To do this, soak the slide in water until the cover glass can be removed, then lift the specimen carefully and transfer it to a new slide. Some technicians find slides easier to prepare if the Hoyer's medium is diluted with water; however, in the process the mounts may collapse as the excess water evaporates. It is strongly recommended that Hoyer's medium be prepared exactly as directed (see Appendix) and used undiluted. Aqueous media are affected by ambient moisture; mounts made in very humid conditions may not dry satisfactorily. Nevertheless, Hoyer's is preferred by most acarologists because its refractive index is excellent for use with mites, and specimens can be mounted directly from the collecting fluid without clearing or fixing. The specimens are cleared after mounting by heating the slides briefly on a hotplate set at 65° C until the medium barely begins to bubble. Do not allow Hoyer's medium to boil or the specimens may be ruined. Such mounts can be prepared quickly for immediate study but should be placed in an oven for curing. (See item (7).)

(5) To place specimens in the medium, put one or more drops of the medium in the center of a 2.5- by 7.5-cm clean glass slide. The precise amount of medium to use will require some experience. Enough is needed to run under the entire cover glass. When Euparal is used, a little more is required than with some other media because of shrinkage, but an excess of any medium around the edge of the cover glass is undesirable.

Place the cleared and washed (also stained or bleached if necessary) specimen in the medium on the slide and make sure that it is well immersed and that air bubbles are absent. Arrange it in the desired position with a fine needle. If the specimen is thick, place at least three pieces of broken cover glass or plastic sheet around it to prevent undue crushing when the cover glass is applied. With some preparations, as for example with ovipositors of tephritid flies, a considerable amount of pressure during drying is desirable to obtain maximally flattened and comparable preparations. Then gently lower a cover glass onto the specimen with forceps, holding the cover glass at a slight angle so that it touches the medium first at one side to prevent air entrapment as much as possible. Apply gentle pressure with the forceps to fix the position of the specimen.

It is often advisable to prepare specimens in more than one position, for example, dorsal side up as well as dorsal side down, but do not mount parts of more than one individual specimen on one slide, because all individuals in the series may not be taxonomically identical.

- (6) Ringing. Special compounds are available to apply in a circle around the edge of the cover glass and the adjacent area of the slide to seal the medium. This is advisable with aqueous media and any that do not harden as they dry. It is not necessary to ring Canada balsam or Euparal mounts.
- (7) Curing. Allow slides to dry or set completely before handling or placing them in other than a horizontal position. Until dry, avoid storing them in a slide box, mailing them, or allowing other persons to use them. If an oven is available, set it at about 45° C and leave the slides in it for at least 24 hours and preferably for several days, up to 3 or 4 weeks.
- (8) Labeling Slides. Collection data should accompany specimens at all times during preparation, either complete data or code symbols referring thereto. Square labels are obtainable from biological supply houses. Excellent ones are now available with pressure-sensitive cement that prevents the labels from popping off the slides as moistened ones often do. Some workers place all information on one label; others use two labels, one at each end of the slide, with the identification on one label and the collection data on the other. All data should be written clearly with India ink, typeset, or typed and reproduced photographically. An effective and flexible method for labeling slides is to use a memory typewriter with small (microgothic) type. The typewriter will automatically repeat as many labels as desired for the same species and collection data. The kind of mounting medium used should also be noted on a label if remounting is necessary.

Sample Procedures

The following procedures have been successful for general use by specialists in the Systematic Entomology Laboratory. Dissecting, staining, and mounting Lepidoptera genitalia are highly specialized procedures that are not included here. Many other procedures are well adapted for general use, but the simplicity and dependability of the following make them preferred by many specialists.

Preparation and Storage of Genitalia. The structures at the end of the insect abdomen in both sexes are the postabdomen, terminalia, or genitalia, although the last term is more restrictive and refers morphologically only to certain organs of the ninth abdominal segment. These structures, sometimes extending to modifications of many segments of the abdomen, are of great identification importance. Many insects cannot be identified to species without critical examination of these parts, and even then can only be identified in one sex. In some insects, these parts are seen easily without special preparation; in others, just a little special positioning of the genitalia at the time the insects are pinned is sufficient. But in many insect species, these structures are

so withdrawn or folded that, for critical examination, the abdomen or a large part of it must be removed and the genitalia specially prepared as follows:

- (1) Carefully remove the abdomen by grasping it with forceps as close as possible to the thorax. Bending it slightly upward, then downward, will usually break it free of the specimen. It is well to perform this operation over a small dissecting dish containing water or 70 percent ethanol into which the part can fall. If the specimen is in a fluid and therefore soft, the abdomen may be severed with fine scissors.
- (2) Place the severed abdomen in a small beaker or crucible containing three pellets of sodium hydroxide (NaOH) in about 10 ml of water. Then set the container on a hotplate at a temperature a little above that needed to boil the solution. It is well to place a cover loosely over the container to prevent the specimen from being thrown out and lost if the solution 'bumps' when heating, or to use a copper-mesh screen between the hotplate and crucible to eliminate 'bumping.' Allow the solution with the specimen in it to boil for 1 minute. Great care must be taken to avoid getting hot or cold NaOH or caustic on any part of the person. If that should happen, wash it off immediately with plenty of water.
- (3) Remove the specimen with forceps and return it to the dissecting dish. Examine it to see that muscles and most internal organs have been dissolved. If not completely so, return the specimen to the solution and heat it a little more.
- (4a) When the specimen is well macerated, take a pair of No. 1 stainless steel insect pins, glued in wooden handles with a drop of epoxy, and pry the genitalia into an extended position. Clear away unwanted parts or debris, or if much unwanted material is present, transfer the specimen to a clean dish of 70 percent ethanol. Water or dilute alcohol is better than glycerin in which to examine small, colorless specimens, partly because fine structures are more clearly visible. The water may be tapwater, but it should be boiled before use to remove dissolved gases that may collect on and in the specimen and be very difficult to remove. The specimen then may be examined and identified or, if necessary, it may be placed in an aqueous solution of a few grains of dry Mercurochrome for staining (see p. 36). The specimen may be held at various angles with a bent piece of minuten and even sketched. If it is to be preserved for permanent reference, the decision must be made whether to store it in a microvial or to mount it on a microscope slide.
- (4b) Another useful method when several specimens are to be identified simultaneously includes using a 'spot,' 'well,' or 'depression' plate, which is a white or black ceramic dish with generally 12 wells on the surface. The same number of wells are utilized as the

number of specimens to be identified. (When using more than one specimen, be absolutely certain that the abdomens are properly associated with the correct specimens. To insure this, place the abdomens in the wells in the same order or configuration as the specimens are arranged in their holding container, and mark one side of the plate to indicate its orientation.) To each well add water and one pellet of NaOH with forceps. After the NaOH has dissolved, place one abdomen or part of it in each depression, and warm the plate gently under an incandescent bulb for about 1 hour. After some of the water has evaporated, replace it with fresh distilled water. Also at this time, examine the abdomens and press out any large air bubbles trapped within that might prevent penetration of the caustic. Then reposition the plate under the bulb. A thin stream of macerated tissues soon will be seen to issue from the abdomens into the fresh solution. After an hour or so, depending on the degree of maceration desired. transfer the abdomens for a few minutes to the wells of a second plate that you have filled with 70-80 percent alcohol to which has been added a small amount of acetic acid to neutralize the caustic. While in these wells, the abdomens may be gently manipulated to remove any remaining tissues. Wash and dry the first plate, place 2 drops of glycerin in each well, top with 70-80 percent alcohol, and transfer the abdomens to this plate, using care to keep them in the proper order. The abdomens may now be examined or left in a clean open place for several days if necessary. The glycerin will not evaporate. If the genitalia are to be permanently preserved, place the parts in a microvial as described on page 39.

Mount the specimen on a microscope slide only if it is relatively flat and all needed characters can be seen in the final position. For example, the ovipositors of fruit flies (Tephritidae) are flat enough that they may be fully extended and the ovipositor and sheath, including spermathecae, can be mounted on a slide with all necessary characters well displayed. The postabdomens of the male tephritids, however, are ill suited to such treatment because they are about as thick as they are wide and must be examined in profile as well as in ventral and posterior view.

(5a) If the specimen is to be mounted on a slide, place it in a small dish of 95 percent ethanol for a short time (1 minute is usually sufficient), then add a drop or more as needed of Euparal on a slide. Remove the specimen from the ethanol and immediately place it in the desired position in the Euparal. Break any large bubbles present before carefully lowering the cover glass. If insufficient Euparal is present to run to the entire circumference of the cover glass, add a little more at the edge of the cover glass until a light pressure on the top of the specimen through the cover glass brings the Euparal to the entire edge. Label the slide and allow it to cure (see p. 37) overnight in a warm oven or for a few days in a clean open place to make it usable. Small bubbles will disappear, and the specimen will become a little more transparent.

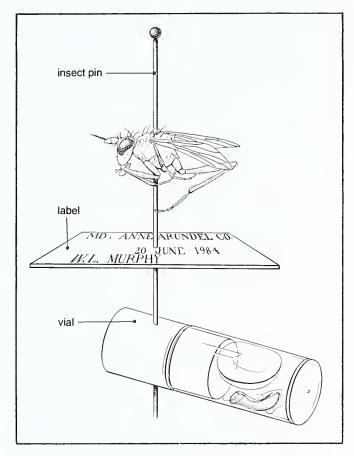


Figure 23. Specimen-microvial mount for storing genitalia preparations on same pin with specimen from which abdomen was removed.

With the aid of an ocular grid in the microscope, the genitalia may be examined and even sketched when lying in a small dish of water, which gives more contrast than glycerin to delicate structures that may be difficult to see. The object may be held in place with a minuten bent in the shape of the letter 'L,' which is laid over it or pierces it at a convenient place. A bit of petroleum jelly will hold a preparation in place, but the jelly must be dissolved before the specimen is replaced in a microvial or mounted on a slide.

(5b) If the specimen is not suitable for mounting on a slide, it may be kept in a microvial. The best microvials are made of transparent plastic with neoprene stoppers. Those with an inner lip are particularly desirable. The former practice was to use glass microvials with cork stoppers, but the tannin in the cork is injurious both to the specimen and, when wet with the glycerin in which the specimen is kept, to the pin on which the preparation is held. Whatever kind of microvial is used, before placing the specimen in the vial, add just enough glycerin to the bottom to cover the specimen completely. A throwaway injection syringe is excellent for this purpose. It may be kept filled with enough glycerin for many preparations. A small container of squeezable plastic with a fine tubular nozzle is made for modelmakers to dispense plastic cement. It is also

an excellent glycerin dispenser. After placing the specimen in the vial, add the stopper. A dull-pointed pin inserted between the stopper and vial allows pressure to escape and prevents droppage of the vial from the stopper, which is to be held by an insect pin, preferably the same pin carrying the specimen from which the genitalia preparation was made (fig. 23). The specimen may be removed from the microvial and reexamined in water or ethanol solution at any time and then replaced.

Reference: 143 (slide-mounting genitalia of Lepidoptera).

Mounting Wings. Wings of many kinds of insects can be mounted on microslides for detailed study or photography. Those covered with scales, such as wings of Lepidoptera and mosquitoes, must first have the scales removed or at least bleached for study of the venation.

Wings are bleached by immersion in an ordinary laundry bleach (sodium hypochlorite solution). Wetting them first with ethanol will activate the bleach. Immersion in the bleach for 1-3 minutes is usually sufficient. As soon as the veins become visible, remove the specimen or part from the bleach and wash it in plain water. It is frequently desirable to remove the scales under water by brushing the wings carefully with a fine brush or with the tip of a small feather. The descaled wing may then be stained, if desired, in eosin-Y or in an aqueous solution of Mercurochrome for a few to several hours and then washed again. The wing is then ready for mounting as described here, or it may be allowed to dry on a slide, then placed under a cover glass, and the cover glass ringed with fingernail polish or ringing compound to hold it in place.

Wings not needing descaling may be removed from a fresh specimen or one that has had a drop of household ammonia (containing detergent) placed at the base of the wing and allowed to stand in a closed receptacle for about an hour. The wing may be removed with finepointed forceps by piercing the body cuticle surrounding the wing base and then pulling the wing loose. In this way, one may be assured of obtaining the complete wing, even with basal sclerites if desired. The wing is then wet with 70 percent ethanol and placed in plain water for about 10 minutes to soften it. It is often desirable, if the wing is from a dried specimen, to place it in water that is then carefully heated until it barely starts to boil. This will aid in removing air from the larger veins. While the wing is in the water, carefully remove any dirt that may be present with a fine brush, but avoid removing fine hairs and setae. Also remove any unwanted parts of body cuticle and muscles at the base of the wing.

Then place the wing for about half a minute in 95 percent ethanol while adding a few drops of Euparal to a slide. Remove the wing from the ethanol and immediately place it in the Euparal on the slide. Position the wing as desired, turning it over if necessary and making sure that its basal part is well stretched out. Alternatively, especially with very delicate wings, it is usually better to arrange the wet wing on the bare slide first, then pour the mounting medium on top. Carefully apply a cover glass, touching it to one side of the Euparal first at a slight angle from horizontal to avoid entrapping bubbles. Press the cover glass down on the wing carefully to expand it as much as possible and to force bubbles out of the basal veins and elsewhere. Then cure the slide in a warm oven overnight or in the open, clean air for a few days.

Mounting Larvae of Diptera, Coleoptera, Lepidoptera, and Other Groups. The study of the immature stages of many insects is of great importance for identification purposes, but special techniques are usually needed because of their soft cuticle. Immature insects of most groups are seldom suitable for preservation in a dry condition. A method given here for preparing dipterous larvae may also be used for immatures of some other groups. Dipterous larvae, especially those of the higher Diptera, have mouthparts, a cephalopharyngeal skeleton, anterior and posterior spiracular structures, anal plates, cuticular spicules, and other features that are important for their systematic study, but these parts usually must be examined at high magnification and require special treatment. The larvae of Diptera, Coleoptera, Lepidoptera, and many other groups are best killed in boiling water because it leaves them in good condition for critical examination.

For cursory examination of the internal cephalopharyngeal skeleton, place the larva with no more fluid than will adhere to it in a dissecting dish. Pierce the cuticle in a few places near the anterior end of the larva and apply a few drops of pure liquid phenol there. Be careful not to get any phenol on your skin; wash with water if you do. In a short time the tissues will become as clear as glass. The larva may be returned to 75 percent ethanol after examination, when the tissues will again become opaque.

For more detailed and permanent preparation of larvae, place the larva in water in a dissecting dish and cut the cuticle with fine dissecting scissors along one side, starting close to the anterior end, passing below the lateral spiracle, and continuing almost to the posterior end. Then place the larva in an NaOH solution and boil as described on page 38. When the larva is well macerated, remove the body contents, almost separate the posterior spiracular area from the remainder of the skin, and pull the cephalopharyngeal skeleton a short way out of the body. Place the skin in 95 percent ethanol while adding a few drops of Euparal on a slide. Then put the skin in the Euparal, opened outward so

that the cephalopharyngeal skeleton with the mouth hooks lies away from the skin and the posterior spiracular area lies with both spiracles upward. Apply the cover glass and carefully press it into place. This should give a clear view under high magnification of the cephalopharyngeal skeleton in lateral view, the anterior spiracles, all structures of dorsum and venter of one side, anal plates, and posterior spiracles. The last, often somewhat domed or on conical protuberances, may be distorted, but the sunray hairs and relationships of one spiracle to the other should be easily observed.

As with the genitalia, the larval skin is sometimes best preserved in glycerin in a microvial.

Other parts of the insect body, such as antennae, legs, and palpi, may be mounted on slides in Euparal in the same manner as described for the genitalia, wings, and larvae.

The references cited here concern specialized procedures for making slide mounts of lepidopterous genitalia (180) and methods using Canada balsam (363) for aphids, scale insects, and various other small insects. For methods to use with mites (Acarina), see references on page 52. Further procedures are also given in the Appendix.

References: 180, 363, 463.

Labeling

To have any scientific value, specimens must be accompanied by a label or labels giving, as a very minimum, information about where and when the specimen was collected, who collected it, and, if pertinent, from what host or food plant. During preparation and mounting, specimens should bear temporary labels with this information, and any time a sample is subdivided, the label must be copied so that every specimen continues to be accompanied by the data. Many collectors keep a field notebook to record more detailed information, such as general ecological aspects of the area, abundance and behavior of the specimens, and any other observations noted at the time of collection. A system of code numbers may be used to associate field notes with the specimens collected; however, the code number should appear in addition to the basic data on the label with the specimen. A code number by itself is never a valid permanent label for a specimen.

Paper

The paper used for making labels should be heavy enough so that the labels remain flat and do not rotate loosely on the pin. The surface of the paper should be smooth enough to write on with a fine pen. Linen ledger paper, 100 percent rag and of 36-pound weight, is best. Smooth calendered, two-ply bristolboard is also good; it is

usually obtainable from art supply stores. Also desirable is a heavy, high-rag-content paper, used for professional-grade herbarium sheets; it may be obtained from biological supply houses. Labels made from poor quality paper become yellow and brittle with age, tend to curl, disintegrate in liquid preservatives, and are generally unsatisfactory.

Ink

The ink should be a good grade of India ink that is permanent and will not "run" if the labels are placed in jars or vials of liquid preservative. Be sure the ink is completely dry before placing the label in the liquid. It is also helpful to use a waterproofing spray (artist's fixative) on the labels after they are dry. India ink is not always available when collecting in the field. However, labels written with a firm hand and with a moderately soft lead pencil are satisfactory. Do not use ballpoint pens or hard lead pencils for labels placed in liquids; the writing soon fades and becomes illegible.

Lettered and Printed Labels

Labels may be lettered carefully by hand with a finepointed pen, such as a crow quill. Printed labels, with four-point type, are preferable and are advisable if more than a few of one kind are needed. They may be printed with blank spaces left for the date, or the full data may be printed. Typewritten labels may alternatively be photographed with the proper reduction in size and prints made on a good grade of paper. Typewriters with carbon tape may be used to make a sheet of labels. Then, on a copying machine that can reduce the image size, copies will be clear and legible at the needed reduction, but they cannot be made on sufficiently heavy paper. Therefore, such copies should be firmly cemented with a casein glue to another thin sheet of good paper and well dried under pressure to keep them flat. Then good permanent labels can be cut from the sheets. The newer photo-offset methods can also produce satisfactory labels from typewritten copy, but the proper kind of paper must be specified. Printed labels may also be ordered from a commercial firm, and this is often the most satisfactory approach.

Size of Labels

One must seek a middle ground between the size of the insect on a pin and the amount of data a label will hold. Inasmuch as most insects are small and the amount of necessary data takes up more space than most of the insects, try to make labels of a certain maximum size and use more than one label if more data are needed than can be put on one label. Never use more than one side of a label. The maximum size is about 8 by 18 mm, or, in 4-point type, 5 lines of 5 pica length, or about 13 capital letters; however, commercial labels can be much smaller. Large beetles and butterflies need larger labels, but avoid so-called "barndoor" labels because they do not hold well on a pin. Even with very small insects, do not skimp on the amount of data just to make a small label. One advantage of using moderately large labels with small insects is that if

a pin with such a label is accidentally dropped, the label will often keep the insect from being damaged. If capital and lowercase letters are used, it is not necessary to use spaces between words, as JBSmith, NewYork, LittleFalls. If there is any chance of ambiguity, it is best to use full spellings if there is sufficient room. With only one line of data, the label should be wide enough so that when the pin is inserted, all data are legible.

Label Data

The indispensable data must answer the questions of where, when, and who, in that order and as exactly as feasible. Only the size of the label should limit the amount of data. This kind of data, usually known as locality, date, and collector data, should be given as follows:

- (1) Locality. The collection locality should be given in such a manner that it can be found on any good map. If the place is not an officially named locality, it should be given in terms of approximate direction and distance from such a locality, or the coordinates of latitude and longitude may be given. The Smithsonian Institution (U.S. National Museum) recommends that for localities in the United States and Canada, the name of the State or Province be spelled in capital letters, such as ONTARIO, ALBERTA, MARYLAND, NEW YORK, and SO. CAROLINA. This method should also be used for foreign countries, as ENGLAND, PAKISTAN, GERMANY (WEST), and SRI LANKA. Then, if at all feasible, the next subordinate region should be cited in capitals and lowercase letters, such as counties and parishes in the United States and Canada and provinces elsewhere. Here are a few examples, with a virgule (/) indicating the end of a line: ARIZONA/CochiseCo./15kmNEPearce (=15 km northeast of Pearce); NEWFOUNDLAND/ Hermitage Dist./12kmWStAlbans; EGYPT Cairo/SuezRoad 38kmW/Suez;EGYPT Mud,-AI-/Tahrir22km/SWAbulMatamir; or EGYPT/Mud.-AI-Tahrir/30°05'E,30°15'N. Current twoletter abbreviations for States and zip codes should not be used because they are not self-explanatory and may not be permanent.
- (2) Date. Cite day, month, and year in that order, preferably using the international convention of writing day and year in Arabic numerals and the month in Roman numerals without a line over and under the numerals. It is best to place a period or short dash between each number, for example, 4.VII.1978 (= July 4, 1978), 5.V.1909, 5-V-1909. If a few consecutive days have been spent collecting in one locality but not more than a week, the extreme days may be cited, for example, 5-9.V.1909; or if 3 consecutive nights of light trapping were at one spot, the median day may be cited, as 8.VIII.1984 for trapping done on the nights of the 7th to 9th of August 1984. For reared specimens, the dates of collection of the immature stages and of adult emergence should be cited, as pupa 10.VI.1980, em.24.III.1981, indicating that the pupa was collected on 10 June 1980 and the adult emerged on 24 March 1981.

- (3) Collector. Spell the last name of the collector or collectors, using initials for given names if space permits. If the last name is a common one, such as Smith, Jones, or Williams, always include initials, and if a group with more than three collectors, use the leader's name followed by et al.
- (4) Other Data. It is especially important to cite hosts of parasites and plant-feeding insects when known. Details of the habitat, such as elevation, ecological type, and conditions of collection, are all important and are usually put on a label in addition to the primary data. Such data are "swept from Salsola kali," "Malaise trap, reared ex human feces," "McPhail trap in orange grove," "at light," "3,200 m," "sandy beach," and "under bark dead Populus deltoides." Do not use vernacular names of hosts unless the host is common and widespread, such as orange or horse. If the specific name of a host is not known, at least give the genus. "Vaccinium sp." is better than no name or "huckleberry." Even the family name of the host is helpful if no more specific name is available. The presumed nature of the association between insect and plant should be clearly indicated, for example, "Resting on flowers of Vaccinium sp." The word "ex" (Latin for "out of") should mean that the insect was observed feeding on or in or was bred from the mentioned plant.

As noted earlier, it is advisable to keep a notebook, in which details of locality, habitat, and other important data are kept. Each individual locality may be assigned a notebook or code number with which the collecting jars and vials are marked until the specimens can be prepared, but citation of such a number on permanent labels is virtually useless. Even when large files of such data are kept, they are seldom available many years later when a researcher needs to know what a cited notebook number means.

Placing the Labels

For double-mounted insects, insert the pin through the center of the right side of the label (fig. 24), with the long axis of the label oriented in the same direction as the card point. Use care that the pin is not inserted through, and thereby obscuring, the writing on the label. For specimens mounted by direct pinning, the label is centered under the specimen with the long axis of the label coinciding with the long axis of the specimen. The left margin of the label is toward the head of the insect. An exception to this is when specimens have the wings spread, such as Lepidoptera. The label is always alined transversely, at right angles to the axis of the body, with the upper margin toward the head. Labels may be moved up the pin to the desired height by using a pinning block (fig. 19). The middle step of the block will give about the right height if only one label is used. When more than one label is used, space the labels on the pin beneath the specimen so that the information on the labels can be read without having to move any of them.

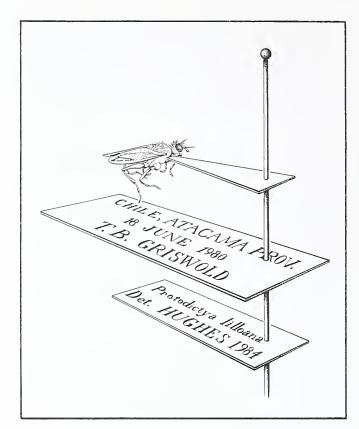


Figure 24. Correct position in which to place labels for double-mounted insects.

Labeling Vials

Material in fluid should be accompanied by a single label large enough to include all data. The label should be written with a moderately soft lead pencil or in India ink and well dried so that it will not dissolve or run when immersed in the liquid. Do not use a ballpoint or felt-tip pen. Hard lead pencil writing becomes illegible in liquid. Do not fold the label. Small specimens may be damaged or lost when the label is removed. Multiple labels or labels small enough to float around in the vial may also damage specimens, and when two labels lie face to face, they cannot be read. Always place labels inside the vial as there is the danger that if left outside a vial, regardless of the method or substance used to affix them, they may become defaced, destroyed, or detached.

Labeling Microscope Slides

To label microscope slides, use square labels made expressly for this purpose and obtainable from biological supply houses. Labels with pressure-sensitive cement are now available. They are far superior to the older labels, which often came off. Put as much data on the label as feasible, including the kind of mounting medium used in case remounting is needed. Many workers use a label on each side, reserving one for the species determination. Never put labels on the underside of a slide.

Identification Labels

When specimens are sent to an expert for identification, they should be accompanied by permanent collection labels giving all essential data. If associated field notes are available, copies of these should accompany the specimens. When the identification has been made, the scientific name of the specimen and the name of the identifier should be printed on a label associated with the specimen. On pinned specimens, this information is always printed on a separate label placed below the collection label or labels on the same pin. When a series of specimens consists of the same species, the identification label is often placed only on the first specimen in the series, with the understanding that all other specimens to the right in that row and in following rows belong to the same species. The series ends with another specimen bearing an identification label. Identifications for specimens preserved in alcohol or on slides may be written on the same label as the collection data or on a separate label, depending on the preference of the collector or person making the identification.

Care of the Collection

If care is taken and a few basic precautions are followed, a collection of insects or mites can be maintained indefinitely. The information given here is general; institutions and individuals will want to adapt materials and procedures to fit their own needs and resources.

Housing the Collection

The adoption of standard equipment for housing a collection is advantageous as it assures uniformity of containers when additions are necessary. Standard equipment is obtainable from any of several supply houses.

Material preserved in liquid usually needs no attention other than occasional replacement of preservative and stoppers. Small vials may be stored in racks so that the stoppers are not in contact with the liquid. The use of storage racks for vials expedites rearrangement and examination of the material. Vials should be examined periodically to be sure the specimens do not become dry. If it is not possible to inspect the vials frequently, those containing larvae or large insects should have their stoppers replaced by cotton plugs. Several such vials can be placed upside down in a single large jar filled with preservative. Use of cotton plugs is not recommended for very small or delicate specimens because they may become entangled in the cotton fibers. Jars with screw tops or clamping lids, as are used in home canning, are ideal, but jars specifically designed for museum use can be obtained from biological supply houses. Stoppers of neoprene or other synthetic materials generally are superior to cork stoppers, but good quality cork stoppers are usually preferred to plastic screw tops, which often are easily broken. Many of the newer flanged plastic stoppers are excellent.

Microscope slides are usually stored in wooden or plastic boxes obtainable from biological supply houses. The inner sides of the boxes are slotted to hold the slides vertically and to separate them from one another. Slide boxes are available in sizes made to hold from 50 to 100 or more slides. If the slides are to be stored vertically, it is important that they be thoroughly cured before storage or the cover glasses may slip. Some workers store the slide boxes on their sides so that the slides rest horizontally. This is especially desirable if the slides are made with Hoyer's medium. which may become soft under very humid conditions. Several slide-filing systems are available from suppliers, but whatever system is used, care should be taken to assure that additional similar equipment will be available in the future for expansion of the collection.

Small plastic slide boxes, usually made to hold five slides, are convenient for keeping slides in a unit-tray system along with pinned specimens. This is especially desirable when genitalia are mounted on slides, because it is readily apparent to visiting researchers examining the pinned specimens that such slides are available.

Pinned specimens are best kept in one of the types of standard, commercially available insect drawers, available in U.S. National Museum, California Academy of Sciences, Cornell, or Schmitt sizes. Larger collections usually use the unit-tray system, with various sizes of unit trays made to fit into a drawer. The pinning bottoms of both the unit trays and boxes are now generally made of polyethylene foam. The older standard was pressed cork, but that was extremely variable in quality and usually contained enough tannin to corrode pins and eventually to cement the pins firmly into the pinning material. Polyethylene foam is now available in large sheets to be cut to the desired size and cemented into boxes or unit trays.

A serviceable substitute for polyethylene is 6-mm-thick balsa wood boards, obtainable from modelmaker supply houses. These boards should be individually selected for softness because they are frequently excessively hard. Another good substitute, especially for temporary storage of pinned specimens, is double-thickness corrugated board, which is often used to separate layers or rows of cans in cartons. Single-thickness corrugated board will not hold an insect pin firmly, and the harder board used for making cartons is not usable.

Any box used to store insect specimens must be nearly airtight to keep out museum pests—dermestid beetles, psocids (booklice), and certain other insects—which will quickly devour or at least make a shambles of a collection. These pests find their way even into the best boxes or insect drawers, and constant vigilance is necessary.

Protecting Specimens From Pests and Mold

Fumigation of all insect storage boxes is necessary. The best made insect drawers provide for chemical fumigants. Two of the most widely used fumigants are paradichlorobenzene (PDB) and naphthalene, both of which are obtainable in balls or flakes. Never mix PDB with naphthalene as they react chemically and produce a liquid that may damage the collection.

Solid fumigants should not be placed loose in a box of pinned specimens. If crystals or flakes are used, a small quantity should be placed in a little cloth bag or in a pillbox with the top perforated with tiny holes. This container is pinned firmly into one corner of the box of specimens. Mothballs may be pinned in a box by attaching the mothball to the head of an ordinary pin. This is done by heating the pin and forcing its head into the mothball. When moving boxes, be careful that the mothballs and fumigant containers do not come loose and damage the specimens.

To kill pests that are actively damaging a collection, you may need to use a liquid fumigant, which acts more rapidly than solid fumigants. Examples of liquid fumigants are carbon disulfide, carbon tetrachloride, chloroform, ethyl acetate, and ethylene dichloride. Because liquid fumigants volatilize rapidly, may be flammable, and are *toxic to humans*, use extreme care. Work outdoors if possible and use some kind of fumigation chamber. A large plastic bag will serve this purpose. A cotton ball, saturated with a liquid fumigant, is placed in the infested box, which in turn is placed in the fumigation chamber or plastic bag. One day in the chamber usually is sufficient to kill the pests.

Another useful method to kill pests in a collection is to cut strips of dichlorvos into small pieces and place them in the insect drawers. This method gives a fairly rapid kill while avoiding the hazards of using flammable liquids. To keep museum pests out of Riker mounts and other display cases, sprinkle naphthalene flakes on the cotton when the mount is prepared. Papered specimens should be kept in boxes with PDB or naphthalene.

All fumigants are toxic to humans to some extent, and most of them are highly flammable. Even PDB, commonly sold for household use, is now considered toxic to some degree. Before using any fumigant, it is well to find out as much as possible about its properties.

Another serious problem with insect collections, especially in moist, warm climates, is mold, a kind of fungus that readily attacks and grows on insect specimens. Once a specimen has become moldy, nothing can be done to restore it. If only a few filaments or hyphae of mold are present on a specimen, they may be removed carefully with forceps or with a fine brush and the specimen dried well in a warm oven and then restored to the collection. Only keeping the collection

in a dry place will prevent mold. In humid climates it is sometimes necessary to keep insect and other kinds of collections in rooms with artificial dehydration. Some microscope-slide mounting media are also subject to molding.

Reference: 243.

Packing and Shipping Specimens

In mailing insects and mites, there is always a risk of damaging or losing specimens. By following the recommendations given here, the risk can be reduced to a minimum.

Packing Materials. Cartons may be of strong corrugated board or other stiff material; wood is advisable for overseas shipments. Screw-top mailing tubes are good for small items. All containers must be large enough to include ample packing material to minimize the effects of jarring—a minimum of 5 cm on all sides. The packing material may be excelsior or shaved wood, crumpled newspapers, or foam plastic bits. One of the best materials is the clear plastic sheet material with a regular pattern of bubbles (bubble pack or blister pack). This is very light weight and has excellent shock-deadening properties.

Pinned Specimens. Pinned specimens should always be placed as described here in a small box with a pinning bottom. The box should be well wrapped and placed in a larger carton with at least 5 cm of lightly packed packing material between it and the carton on all sides.

- (1) Use a sturdy pinning box with a good pinning bottom at least 6 mm thick cemented securely to the bottom of the box. The box should have a tight lid or one held in place with a strip of masking tape. Do not mail specimens in an open-top museum tray.
- (2) Pin the specimens firmly into the pinning bottom, leaving enough space for easy removal. Place bracing pins on each side of heavy or long-bodied specimens to prevent them from rotating (cartwheeling) on their pins and damaging adjacent specimens. Microvials should have an additional pin at the end of the vial to keep it from coming off its stopper. Vials other than microvials should be wrapped in a box separate from pinned specimens.
- (3) Unless the box in which the specimens are pinned is shallow enough so that the heads of the pins almost touch the lid, a piece of firm cardboard should be cut to fit into the box and lie on top of the pins. If there are only a few specimens in the box, a few extra pins should be added near the corners to keep the cardboard level. It is helpful to attach a tab made of a piece of adhesive tape folded double, with the ends left free to attach to the top of the inserted cardboard. The insert may be lifted out by the tab. The space between the insert and the lid of the box should be filled with enough packing material, preferably cotton batting, not

excelsior or any shredded or loose material, to keep the insert pressed lightly against the tops of the pins when the lid is in place. This prevents the pins from working loose and wreaking havoc in transit.

(4) If only one or two specimens are being shipped, they may be placed in a straight-sided plastic vial with a press-on or screw-on top. The vial should be of sufficient diameter to hold the labels in a normal position. A cork stopper cut to such a length that its larger end is a little greater in diameter than that of the inside of the vial is pressed tightly into the bottom of the vial. This will provide a good pinning bottom into which one or two pinned specimens may be firmly pressed. Attach the cover of the vial, wrap the vial in enough packing material to hold it firmly in a mailing tube, attach the cover of the mailing tube, and it is ready to ship.

Although it is good practice to fumigate boxes before shipping, do not leave loose fumigant in the box with the specimens nor any fumigant balls on pins in containers. They are especially prone to work loose and damage specimens.

Specimens in Vials. The following procedures are recommended:

- (1) Fill each vial with liquid preservative. Stopper tightly by holding a pin or piece of wire between the vial and the stopper to permit air or excess fluid to escape, then remove the pin or wire. Make certain that cork stoppers do not have defects that will allow leakage. Screw-top vials should be firmly closed and sealed with a turn and a half of plastic adhesive tape around the lower edge of the cap and part of the vial. There is no need to seal with paraffin; it often breaks loose and will not prevent leakage.
- (2) Wrap each vial with cotton, tissue, paper toweling, or similar material. Allow no piece of glass to come into contact with another piece of glass. Several vials may be wrapped together or held with tape or rubberbands as a unit, or they may be placed in a small cardboard box with enough packing to insure that they are not shaken around.

Loading Cartons. After pinned specimens, specimens in vials, or both have been prepared properly, they should be placed in a strong carton large enough to hold at least 5 cm of packing material around all sides, including the top and bottom. Use enough packing material to prevent the contents of the carton from moving about, but do not pack the material tightly. It should be resilient enough to absorb shocks and prevent damage to the contents being shipped. One or a few vials may be shipped in a mailing tube as previously described. When shipping more than one box or packet of vials, tie or wrap them together as a unit before placing them in the larger carton. Individual boxes or vials otherwise may easily be overlooked and lost when unpacked. Since vials of specimens in fluid are much heavier than boxes

of pinned specimens, cartons containing many vials may be packed somewhat tighter in the carton than those containing only pinned specimens since they tend to remain relatively stationary in the carton. It is not necessary to ship pinned and liquid-preserved specimens in separate cartons, but if there are many of the latter, it is advisable to ship them separately.

Shipping Microscope Slides. First of all, make certain that any slide shipped is thoroughly dried and cured. Slides may be shipped in holders made expressly for that purpose and available commercially from biological supply houses. Even in these holders it is advisable to wrap a little soft tissue around each end of each slide so that the cover glass does not come into contact with anything. The slides may also be shipped in standard storage boxes with enough soft tissue around each end of each slide and between the slides and the box lid to prevent movement. The box should then be wrapped to hold the lid down firmly. It may then be treated as described here for pinned and liquidpreserved specimens. The wrapped slide container may also be tied together with units of pinned or liquidpreserved specimens or both and placed in a carton with them. If no slide holders are available, a few slides, each wrapped with tissue, may be tied together at each end with tape, rubberbands, or string, wrapped in strong paper, and placed in a mailing tube or carton with packing material.

Shipping Live Specimens. Most insects and mites intended for a collection or submitted to experts for identification should not be shipped alive. To protect American agriculture, Federal law prohibits the importation and movement of live pests, pathogens, vectors, and articles that might harbor these organisms unless the shipments are authorized by the U.S. Department of Agriculture. If it is necessary to ship live material, be sure to comply with all Federal, State, and local regulations (sample form is in Appendix). Shipments of live insect material without valid permits may be seized and destroyed by plant quarantine inspectors. In addition to meeting Federal laws, the shipment of some species must be approved by State officials. To obtain Federal authorization, write well in advance of shipment to Animal and Plant Health Inspection Service, USDA, Federal Building, Hyattsville, Md. 20782. Each request is evaluated according to its individual merits. No permits are required for shipments of dead specimens.

Pupae or larvae shipped to be reared elsewhere should be placed in tightly closed containers without vent holes. These insects require a minimum of air and will not suffocate. Pupae preferably should be packed loosely in moist (not wet) moss or similar material. Larvae should be packed with enough food material to last until their arrival. Most beetle larvae and some caterpillars, especially cutworms, should be isolated, since they are rather cannibalistic. To prevent excessive accumulation of frass (fecal material) and moisture, do

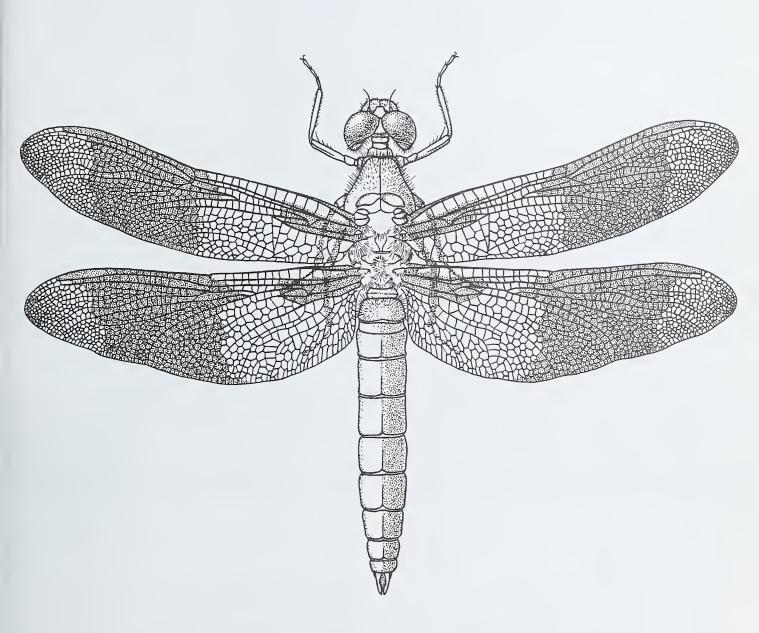
not overload containers. Plant material held without ventilation tends to become moldy, especially when kept in plastic bags. For this reason, pieces of the host plant bearing such insects as scale insects (Coccoidea) should be partially or completely dried before being placed in a container, or they should be packed in a container such as a paper bag, which will permit drying to continue after closure. Live Heteroptera and other small, active insects are killed easily by excessive moisture in the container. Therefore, it is advisable to provide several tiny vent holes or place a fine mesh screen over one end of the container when shipping such insects.

Some containers designed to hold living insects are strong enough to be shipped without additional packing, but generally the containers should be enclosed in a second carton with enough packing material to prevent damage to the inner carton. In all cases, affix a permit for shipping live insects in a conspicuous place on the outside of the shipping container.

There are no restrictions on mailing dead specimens within the United States or to other countries, but if the specimens are to be sent out of the country, U.S. Customs requires that the contents and value of the package be listed. The statement "Dead Insects for Scientific Study, of No Commercial Value" will suffice. It is recommended that all packages be marked "FRAGILE" and that a complete return address be included on the outside of each container.

Part 2

Classification, Biology, and Special Techniques



Classification of Insects and Mites

Classification is essential in any kind of work with organisms. To determine the best collecting methods and techniques for preservation and how best to control pest organisms, identification is necessary, at least as far as the order for even the most elementary study of insects and mites. The information and keys presented here are designed to enable even a beginning collector to recognize the most common orders. The listed references will provide further information on identifying specimens, but because of the immense number of insect and mite species, it is impossible to provide a complete classification either here or in any other single reference.

References (general taxonomic): 46, 47, 60, 80, 89, 117, 118, 207, 221, 224, 225 (mites), 245 (mites), 271, 280 (mites), 291, 330, 333, 337, 344, 370, 434.

Classification is the systematic or taxonomic (taxon: taxa, pl.) arrangement of organisms based on sets of characters common to each group, with each taxon further divided stepwise to species: Kingdom, phylum, class, order, family, genus, species. In many cases, there are also intermediate superdivisions and subdivisions, as well as the less used groups of tribe, cohort, and phalanx. This stepped arrangement is known as a hierarchical system. The kingdom Animalia is divided into a number of phyla, the largest of which is the phylum Arthropoda, containing the insects, mites, and all other animals having an exoskeleton and jointed legs. This phylum is divided into classes as shown in the following key. Additional keys are also given to orders and in a few cases to suborders. For further identification, refer to the vast taxonomic literature.

The keys given here are in the form most often used in American publications. Start at couplet 1. Read the two choices and select the one that best describes your specimen. To the right is a number directing you to another numbered couplet from which you must again choose. Repeat this procedure until a name is reached. A similar procedure may be used with other keys in the literature to which you may wish to refer. The use of keys is far more satisfactory than comparison with pictures or even with identified specimens because the most important characteristics to look for are specifically stated. These are often differences not even to be suspected by an inexperienced person.

It is highly advisable to refer to an elementary textbook on entomology (see the previous references) and become familiar with the basic elements of insect anatomy. Even such words as "face" and "leg" do not mean quite the same when referring to an insect as they do to a human being. It is also important to know how to tell whether the insect you have is an immature or an adult. This kind of information must be found in general textbooks.

Scientific names are used throughout the world. The name of a species is composed of two words—the genus (initial capital) and the species (or epithet, or trivial name; not initial capital). This combination is known as a binomen (plural binomina) and must be written in a typeface different from the text in which it appears, usually in italic. Each genus may be used only once in the animal kindgom; the same species may be used in different genera, but not more than once in a genus. These names are regulated by the International Commission on Zoological Nomenclature, which publishes the "International Code of Zoological Nomenclature." The similar code for plant names differs in some respects from the Zoological Code. There are, of course, common names for insects in all languages. These vary greatly, even in parts of one country, and are not precise. In the United States, certain insects and mites of recognized pest status have common names selected by a committee of the Entomological Society of America and published in "Common Names of Insects & Related Organisms," last revised in 1982. Common insect names ending in words applied to most insects of an order, such as horse fly for certain true flies of the order Diptera, are written as two words, but such names as butterfly and caddisfly for insects belonging to other orders (that is, not true flies) are written as one compound word.

The names of groups above the rank of species are single words, capitalized but not printed in a different typeface from that of text in which they appear. Only the names of families and subfamilies are regulated by the International Code, which specifies that family names must end in "-idae" and subfamily names in "-inae." These endings are added to the stem of the genus name, which is considered typical of the higher taxon, and that generic name is called the basonym of the group name. All group names are plural.

Names above the rank of family are not regulated by the Code and are therefore subject to considerable difference in usage. They are sometimes formed, as are those of families and subfamilies, on a basonym, but many are formed on a word descriptive of a diagnostic character (for example, the name of the order Diptera means two-winged in classical Greek). For these reasons, alternative names of some classes and orders are cited here.

The name of the person who first proposed a scientific name is often appended to the name as a reference; sometimes the year of the proposal is also added after a comma. These references, when added to a binomen, are placed in parentheses if the species name was originally combined with a different genus name, although the current trend is to eliminate the parentheses. This name and year reference is not a necessary part of the binomen, but it is of value to taxonomists in that it sometimes helps verify that different researchers are discussing the same taxon.

Key to Classes of Arthropoda

This key includes all living classes of Arthropoda except Pentastomida (or Linguatulida) and Tardigrada. They are small groups of small to minute, seldom seen animals that lack antennae and are not likely to be taken for Arthropoda. In fact, some authorities do not so consider them.

	 Without antennae (in immature stages, or even in some adults of certain insects, antennae are much reduced or lacking; such forms will not key out 	1.
2	well in this part of the key)	
4	With antennae	
	2. Usually with 7 pairs of appendages, 5 of them legs; abdomen	2.
ADINC	rudimentary (sea spiders; not further considered here)PYCNOG	
	With 6 (rarely fewer) pairs of appendages, 4 (rarely 5) of them legs; abdomen usually well developed as a body region but sometimes fused with	
3	cephalothorax	
	3. Abdomen with booklike gills on underside of abdomen; large animals up to	3.
	50 cm long, with hard expanded shell and long spinelike tail (horseshoe	
	crabs; not further considered here)	
SURA	Class MEROSTOMATA, order or subclass XIPH	
	Abdomen without booklike gills; smaller forms rarely over 7 cm long, body not	
ADINF	as above (spiders, scorpions, mites, etc.; see p. 50)ARAC	
	l. With 2 pairs of antennae (1 pair may be rudimentary in sowbugs); head merged	4.
	with thorax to form cephalothorax; breathing by gills (crabs, lobsters,	
	shrimps, sowbugs, etc.; see p. 53)CRUS	
5	With 1 pair of antennae; head separate from thorax; breathing by tracheae	_
	5. With 3 pairs of legs at some stage in life cycle; body in 3 divisions—head,	5.
	thorax, and abdomen; true legs only on thorax, but abdomen sometimes	
SECTA	with appendages resembling but differing from those on thorax; wings often present (all insects; see p. 54)IN	
SECIA	With 9 or more pairs of legs; most body segments behind head with legs; wings	
6	lacking; body more or less wormlike (myriapodan classes)	
0	5. Most segments of body with double pairs of legs; slow-moving animals	6
PODA	(millipedes; see p. 53)DIPLO	V.
7	Legs not in double pairs; most segments of body with 1 pair	
•	'. Body more or less flattened; not minute, with 15 or more pairs of legs; rapidly	7
PODA	moving animals (centipedes; see p. 53)CHILC	
	Body usually cylindrical; small to minute forms not over 8 mm long, with 9-12	
8	pairs of legs	
	8. Antenna branched; 9 pairs of legs; minute animals 1-1.5 mm long, found in	8.
PODA	leaf litter, etc. (pauropods; not further considered here)PAURO	
	Antenna not branched; 10-12 pairs of legs; white, cylindrical, centipedelike	
HYLA	animals 1-8 mm long (symphylans; see p. 53)SYM	

Class Arachnida

The Arachnida are the largest class of arthropods next to the insects and also contain the next largest number of species important to agriculture (fig. 25). Most arachnids are predaceous; some are parasitic on animals, including humans, but many mites (subclass Acari) are important pests of plants. All arachnids should be collected and preserved in 70-80 percent alcohol. Mites should be treated specially (see p. 95).

References: 90, 368.

Key to Orders of Arachnida

	Abdomen not segmented, or if rarely with distinct plates (sclerites), as in Asiatic family Liphistiidae, also with spinning organs on lower side of abdomen	1.
3 	Abdomen distinctly segmented, without silk-spinning organs on abdomen 2. Abdomen joined to cephalothorax by narrow, short stalk; usually soft and weakly sclerotized, always with spinning organs (spiders; not further considered here)	2.
ACARI	Abdomen broadly fused with cephalothorax (mites and ticks; see p. 52)	
. 4	3. Abdomen with hindsegments forming contrasting, long, taillike prolongation	3.
i i	Abdomen without such prolongation	4.
ı	Abdominal prolongation very slender and many-segmented, not ending in sting; abdomen narrowed to base, without ventral comblike organs	
: I ONIDA)	 Foremost leglike appendages (pedipalpi) slender, similar to legs; minute animals not over 2 mm long, of warm regions (not further considered here)PALPIGRADI (MICROTHELYPHO 	5.
in part)	Pedipalpi very stout, contrasting with very long 1st pair of legs; 2-65 mm long (whip scorpions or vinegaroons; not further considered here)	•
	 Abdomen constricted at base; front legs very long and with long tarsi; pedipalpi clawed at tip; tropical animals 4-45 mm long (tailless whip scorpions; not further considered here) 	6.
7 THIDA)	AMBLYPYGI (SCHIZOMIDA; PEDIPALPIDA) Abdomen broadly joined to cephalothorax; front tarsi not lengthened 7. Pedipalpi with large, pincerlike claws (pseudoscorpions; not further considered here)	7.
	8. Head distinct from 3-segmented thorax; jaws relatively large and powerful, their pincers opening up and down; pale colored, nocturnal, large animals (up to 7 cm long); in Florida and the Southwest (wind scorpions; not further considered here)SOLP	8.
	Cephalothorax not divided into head and 3 segments; jaws usually smaller, pincers not moving up and down	
	 Abdomen apparently 4-segmented, with lateral as well as dorsal plates, and with small, several-segmented endpiece; eyes absent; heavy bodied animals 5-10 mm long, with moderately long legs, occurring as far north as Texas (ricinuleids; not further considered here)RICI 	9.
	Abdomen usually appearing 7-segmented from above, without separate lateral plates; body 5-10 mm long, with legs very long (daddy longlegs, harvestmen; not further considered here)OPILIONES (PHALAN	

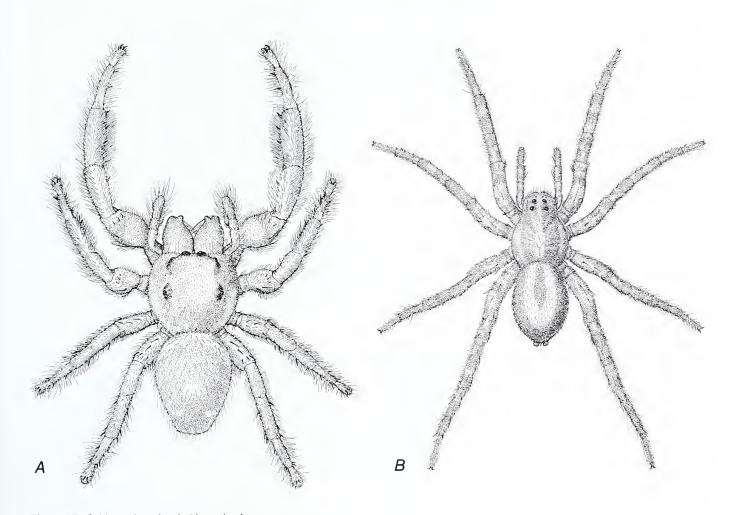


Figure 25. Spiders, class Arachnida, order Araneida: *A*, Jumping spider, *Phidippus audax* (Hentz) (Salticidae); *B*, wolf or ground spider, *Lycosa* sp. (Lycosidae).

Subclass Acari

The acarines are so various in form that their anatomical terminology has developed along considerably different lines than that of the insects. This, together with the fact that most of them are very small and require special techniques for their preservation and examination, has resulted in the group presenting considerably greater difficulty than most insects. Only recently has their higher classification attained a measure of stability.

Previously, the Acarina were generally considered to be an order equivalent to the other orders of the class Arachnida, as Araneida and Scorpionida, but recent students of the mites have been considering the Acarina as the subclass Acari without regard to the status of the other orders. The subclass Acari contains the orders Acariformes and Parasitiformes. These orders, along with the suborders Mesostigmata, Ixodida, Prostigmata, Astigmata, and Cryptostigmata, are used in the classification of mites in the Systematic Entomology Laboratory. Until this difficulty is resolved, the group names used here are left without rank designation.

A few definitions of the more unfamiliar words may be helpful in the following key. A sejugal furrow is a line of demarcation that separates the propodosoma and hysterosoma; a hypostome is the anteroventral region of the gnathosoma (foremost part of the mouthparts); and Haller's organ is a sensory organ found on tarsus I (on ticks only).

of the Subclass Acari

- Key to Some Primary Groups 1. Without visible stigmata (breathing pores) posterior to coxae II; coxae not free, often fused into ventral body wall, forming coxosternal regions delimited by epimera, but sternum lacking; sejugal furrow or interval present, causing legs III to be farther from legs II than the latter are from legs I; number of legs sometimes reduced -----Order ACARIFORMES, including suborders -----PROSTIGMATA, ASTIGMATA, and CRYPTOSTIGMATA With 1-4 pairs of dorsolateral or ventrolateral stigmata posterior to coxa II; coxae free, distinct; sternum nearly always present (lacking in Ixodida); sejugal furrow or interval lacking; distance between legs II and III not greater than between I and II and IV, all of which are present (Order PARASITIFORMES) -----2
 - 2. Pedipalpal tarsus without claws; hypostome modified into piercing organ with backward-directed teeth; stigmata present behind coxa IV or laterad above coxal intervals II to III, each surrounded by stigmal plate; sternum absent; Haller's organ present on upper side of tarsus I (large, bloodsucking acarines known as ticks; length usually well over 2 mm)-----Suborder IXODIDA Pedipalpal tarsus with terminal, subterminal, or basal claw (simple or tined); hypostome serving only as floor of gnathosoma, without teeth; Haller's organ lacking (smaller mites) ---------Suborders MESOSTIGMATA, HOLOTHYRINA, and OPILIOACARIDA

References: 141, 269, 270.

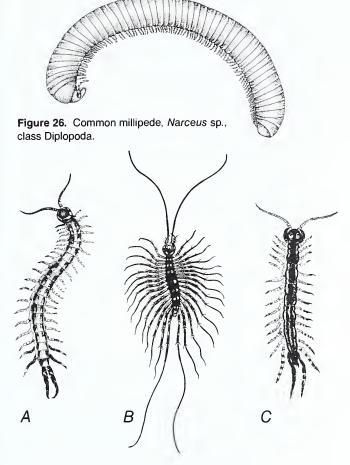


Figure 27. Typical centipedes, class Chilopoda: *A*, Large centipede, *Scolopendra obscura* Newport; *B*, house centipede, *Scutigera coleoptrata* (L.); *C*, small centipede, *Lithobius erythrocephalus* Koch.

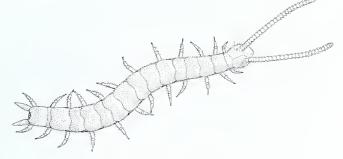


Figure 28. Symphylan, *Scutigerella immaculata* (Newport), class Symphyla.

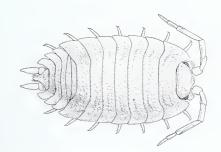


Figure 29. Sowbug, class Crustacea, order Isopoda.

Classes Diplopoda, Chilopoda, Pauropoda, and Symphyla These four classes were once combined as one class, Myriapoda, and are still known as the myriapodan classes. All millipedes (Diplopoda) (fig. 26) are vegetarians, but most do not seriously injure plants. The centipedes (Chilopoda) (fig. 27), however, are predaceous; they possess a pair of strong, poison-supplied jaws to hold and kill their prey. They are fast moving in contrast to the slow-moving millipedes. The minute Pauropoda are of no agricultural importance, but one member of the Symphyla, Scutigerella immaculata (Newport) (fig. 28), is sometimes a pest in greenhouses. It is whitish, about 8 mm long, and may become very numerous.

All members of the myriapodan classes must be killed and preserved in 70-80 percent alcohol. Isopropyl alcohol (isopropanol) seems to have some advantages over ethanol for preserving these animals. Millipedes have the discouraging habit of curling up when dying. The only simple solution to this problem is to remove them from the killing fluid before they stiffen, straighten them out, and then return them to the alcohol in a narrow, straight-sided vial until they become well hardened.

Class Crustacea

The only Crustacea of agricultural importance are the terrestrial isopods known as sowbugs or pillbugs (fig. 29). Many crustaceans, however, are of importance as human food.

The Crustacea are a large class divided into 8 subclasses and about 30 orders. They are largely aquatic and are found especially in saltwater. The sowbugs belong to the subclass Malacostraca and the order Isopoda. They are distinguished by a depressed body with a shield-shaped head and with the other body segments extending to the side. There are seven pairs of walking legs. The two conspicuous antennae are the second pair; the first pair, or antennules, are vestigial. Eggs are carried by the female in a brood sac on the underside of the body. Breathing is by means of paired gills on the lower hindpart of the body. Since the gills must remain moist, sowbugs cannot withstand drying.

Sowbugs are found under stones, logs, and debris on the ground. They feed on vegetable matter and may become pests of tender plants.

Crustaceans may be killed and preserved in 70-80 percent alcohol. Sowbugs may be killed in a cyanide bottle, but they rapidly become dry and brittle.

Class Insecta

The hardened, or sclerotized, parts of the exoskeleton of the Arthropoda do not grow. Consequently, molting or casting-off of the hardened cuticle must take place several times during the insect's period of growth. Each instar or growth stage may change in form to a lesser or greater degree, resulting from a process called metamorphosis. The more primitive groups, the Apterygota and the division Exopterygota of the Pterygota (see following Synopsis of Insect Orders), have a gradual metamorphosis, in which each stage differs relatively little in form from the next, although in many aquatic insects the immature forms may be entirely different from the adult because of the different life habits. The immature stages of insects with gradual (simple or incomplete) metamorphosis are usually called nymphs. However, they are now commonly called larvae (larva, singular), the term formerly restricted to the early stages of insects with greater change from stage to stage, or complete metamorphosis. The wings of insects with incomplete metamorphosis develop externally in the later instars but do not become functional until the adult stage. In one order, the Ephemeroptera, there is even a molt after the wings become functional. The technical term for adult is "imago," and the first adult instar of the Ephemeroptera is called a "subimago."

Insects with complete metamorphosis change greatly in form from one stage to the next, with internal development of wings and including a resting stage, usually immobile, called a pupa (pupae, pl.). The pupal exoskeleton protects the insect during the virtual reorganization of its entire body. The stages in the development of insects with complete metamorphosis are egg, several larval stages, sometimes a prepupal stage, pupa, occasionally subimago, and finally imago or adult.

Because many insects that are wingless (either primarily or secondarily) in the adult stage are difficult to distinguish from immature stages, both the immature and adult insects are treated in the key to the orders (p. 55). Considerable knowledge of the various groups of insects is needed to determine whether a specimen is immature or adult. If functional wings are present, it is certainly an adult (or, if a mayfly, a subimago). Compound eyes, furthermore, are never found in immature insects of the Endopterygota.

The classification of the insects into orders (ordinal classification) adopted here is a conservative one used in the more recent textbooks. Recent studies on various aspects of insect morphology and physiology have led to the proposal of considerably different classifications, but only time and further study will determine their acceptance or rejection. The following synopsis of the classification includes some synonyms and most of the common English vernacular names for members of the various orders, along with a key to these groups.

Synopsis of Insect Orders³

Subclass Apterygota—primitive wingless insects

- (1) Protura (Myrientomata)—proturans
- (2) Thysanura—bristletails, silverfish
- (3) Diplura (Aptera)-diplurans
- (4) Collembola—springtails

Subclass Pterygota—winged and secondarily wingless insects

Division Exopterygota—insects with simple metamorphosis; wings developing externally

- (5) Ephemeroptera (Ephemerida, Plectoptera) mayflies
- (6) Odonata—dragonflies and damselflies
- (7) Orthoptera—grasshoppers, locusts, katydids, crickets

The following four orders were for a long time included with the Orthoptera, and until recently the Blattodea and Mantodea were considered suborders of an order called Dictyoptera.

- (8) Blattodea-cockroaches
- (9) Mantodea—mantids
- (10) Phasmatodea (Phasmatoptera, Phasmoidea, Phasmida)—stick insects, leaf insects, walkingsticks
- (11) Grylloblattodea (Notoptera)—rock crawlers
- (12) Dermaptera (Euplexoptera)—earwigs
- (13) Isoptera—termites
- (14) Embioptera (Embiidina)—webspinners
- (15) Plecoptera-stoneflies
- (16) Psocoptera (Corrodentia)—psocids, booklice
- (17) Zoraptera—zorapterans
- (18) Mallophaga (Phthiriaptera in part)—chewing lice, including bird lice
- (19) Anoplura (Siphunculata; Phthiriaptera in part)—sucking lice
- (20) Thysanoptera—thrips

The following two orders have sometimes been considered suborders of a single order, Hemiptera.

- (21) Homoptera—aphids, cicadas, hoppers (leafhoppers, treehoppers), psyllids, scale insects, whiteflies, and others
- (22) Heteroptera-bugs

Division Endopterygota—insects with complete metamorphosis; wings developing internally

- (23) Coleoptera-beetles, weevils
- (24) Strepsiptera (sometimes included in Coleptera)—twisted-winged parasites
- (25) Mecoptera—scorpionflies, hangingflies
- (26) Neuroptera—neuropterans
- (26a) Suborder Megaloptera (Sialodea)—alderflies, dobsonflies, fishflies, hellgrammites
- (26b) Suborder Raphidiodea—snakeflies

³Includes suborders sometimes considered as orders.

- (26c) Suborder Planipennia (Neuroptera in narrow sense)—antlions, dustywings, lacewings, mantidflies, owlflies, spongillaflies
- (27) Trichoptera—caddisflies
- (28) Lepidoptera—butterflies and moths
- (29) Diptera—"true" flies, mosquitoes, gnats, midges
- (30) Siphonaptera-fleas
- (31) Hymenoptera—ants, bees, wasps, ichneumonflies, sawflies

Key to Insect Orders4

In any comprehensive survey of the larger orders, there are many exceptions, aberrant forms, and so forth that do not fit well in a brief key such as this. It is impractical to include all such forms, especially when including known immature forms, and some orders must appear more than once in the key. With specimens that do not key out satisfactorily here, some of the references on pages 80-93 or an experienced systematic entomologist should be consulted. This is especially true of pupal forms of Trichoptera, Mecoptera, and some Neuroptera.

1.		2
2.	Wings absent or unsuitable for flight (wingless adults and immature stages) Forewings (on mesothorax) wholly or partly horny, leathery, or otherwise strongly differing from wholly membranous hindwings; hindwings	4
	sometimes lacking	3
2	Forewings wholly membranous, or so at least at base	2
٥.	Forewings (wing covers, elytra) uniformly horny, without apparent veins; hindwings, if present, folded both lengthwise and crosswise, hidden under forewings when at rest; mouth with mandibles	4
	g , and , and an	5
4.	End of abdomen with heavy, forcepslike cerci; wings short, leaving most of	
	abdomen exposed; hindwings very delicate, almost circular, radially folded	
	DERMAPTERA, p. 64 Without such cerci; wings usually covering most of abdomen, or if forewings	4
	short, then hindwings elongate, but sometimes absentCOLEOPTERA, p. 7	0
5.		6
	meanifer to mitted to enouning, minimum and the minimum great may e	7
6.	Forewings hardened or leathery basally, with membranous apical part, usually lying flat on abdomen with apical part overlapping; beak arising from front part of headHETEROPTERA, p. 69	9
	Forewings usually wholly membranous or very nearly so, held more or less rooflike over abdomen; beak arising from hindpart of head, projecting downward between forelegsHOMOPTERA, p. 6	
7	Hindwings not folded, similar to forewings; both wings with thickened, very	9
٠.	short basal part separated from rest of wing by suture, so that most of wing	
	can easily be broken off; social insects living in coloniesISOPTERA, p. 68	5
	Hindwings folded fanwise, broader than forewings; wings without breaking	0
Ω	suture { Minute insects, usually less than 6 mm long; forewings small, clublike;	8
Ο.	antennae short, with few segments; parasites of other insects	1
	Usually large or moderately large insects; forewings usually flat and long; antennae usually lengthened and slender, many-segmented; orthopteran	
	orders	9

⁴Includes suborders sometimes considered as orders.

9.	Hindfemora enlarged, modified for jumpingORTHOPTERA, p	
40	Hindfemora not enlarged, similar to other legs	10
10.	Cerci short, unsegmented; body usually elongate and slender (sticklike)	64
	Cerci long or short but segmented; body usually not sticklike	11
11.	Shape oval; all legs similar, adapted to walkingBLATTODEA, p	
	Shape elongate; forelegs raptorial (with spines and modified for grasping prey)	
	MANTODEA. r	. 63
12.	With but 2 well-developed wings, forepair functional, hindpair not winglike and	
	sometimes small and clublike	13
	With 4 wings, hindpair sometimes small but flat or straplike, not clublike	15
13.	Mouthparts forming a sucking or lapping proboscis, rarely rudimentary or virtually lacking; hindwings replaced by clublike halteres; abdomen without	
		76
	tail filamentsDIPTERA, p Mouthparts not functional; hindwings not formed into clublike halteres;	,, , 0
	abdomen with tail filaments	14
14.	Without halteres; antennae inconspicuous, with small scape and pedicel and	
	bristlelike flagellum; forewings with numerous crossveins (few mayflies)	
	EPHEMEROPTERA, p	. 61
	With hindwings reduced to halterlike structures; antennae evident,	
	not bristlelike; venation of forewings apparently reduced to 1 forked vein (male scale insects)	60
15	Wings long, narrow, almost veinless, with long marginal fringes; tarsi 1- or	. 09
10.	2-segmented, with swollen tip; mouthparts conical, fitted for piercing and	
	sucking plant tissues (minute insects)THYSANOPTERA, p	.68
	Wings broader; if fringed, fringe not longer than width of wing; veins usually	
	conspicuous and at least 1 crossvein present; tarsi with more than 2	
4.0	segments and tip not swollen	16
16.	Wings, legs, and body at least in part with elongate, flattened scales and often also with hairs; wings hyaline (glasslike) under color pattern formed by	
	scales; mouthparts consisting of tongue (rarely rudimentary) formed of	
	helically coiled tube; mandibles (fitted for chewing) present only in a few	
	families of small moths with wingspread not over 12 mm LEPIDOPTERA, p	. 73
	Wings, legs, and body not covered with scales, although a few scales	
	sometimes present; color pattern of wing involving wing membrane and/or	47
17	hair	17
	than forewings; antennae prominent	18
	Hindwings without plaited anal area, not larger than forewings; antennae	
	often inconspicuous, bristlelike	20
18.	Tarsi 3-segmented; cerci well developed, usually long and many-segmented	
	Tarsi 5-segmented; cerci not prominent	19
19.	Wings with several subcostal crossveins, surface without hairs or scales	13
	NEUROPTERA, suborder MEGALOPTERA, p	. 72
	Wings without subcostal crossveins, surface with hairs or scales	
	TRICHOPTERA, p	. 72
20.	Antennae short, bristlelike; wings with numerous crossveins forming overall	21
	network; mouthparts with mandibles close to eyes Antennae larger or wings with few crossveins or mouthparts at end of beak	22
21.	Hindwings much smaller than forewings; abdomen with long tail filaments	
	EPHEMEROPTERA, p	. 61
	Hindwings very similar to forewings; abdomen without long tail filaments	
20	Head extended into back with mandibles at and hindwings not folded; wings	.62
22.	Head extended into beak with mandibles at end; hindwings not folded; wings usually with color pattern and numerous crossveins; male genitalia usually	
	swollen, turned forward, and with strong pair of forcepsMECOPTERA, p	. 71
	Head not extended into beak; male genitalia without conspicuous forceps	23
23.	Mouthparts (sometimes lacking) consisting of proboscis without chewing	
	mandibles; cerci lacking; wings with few crossveins	24
	Mouthparts including mandibles fitted for chewing	26

24.	Wings covered with scales forming color pattern; antennae many-segmented; mouthparts (when present) consisting of helically coiled haustellum (tongue)LEPIDOPTERA,	p.	73
	Wings not covered with scales; antennae with few segments; mouthparts consisting of segmented piercing beak		25
	Beak arising from anterior part of headHETEROPTERA, Beak arising from posterior part of head, extended downward between forelegsHOMOPTERA,		
26.	Body and wings covered with whitish powder; wings bordered anteriorly by very narrow cell without row of crossveins; insects less than 5 mm longNEUROPTERA, suborder PLANIPENNIA (Coniopterygidae),	•	
27.	Body and wings not covered with whitish powder; otherwise differing Tarsi 5-segmented		27 28
28	Tarsi with 4 or fewer segments Prothorax fused with mesothorax; hindwings smaller than forewings, latter		31
20.	with no more than 20 cells; abdomen often constricted at base	p.	78
	Prothorax more or less free, sometimes long; forewings and hindwings approximately equal in size, with more than 20 cells		29
29.	Prothorax much longer than head, cylindrical; forelegs similar to others, not enlargedNEUROPTERA, suborder RAPHIDIODEA, Prothorax not longer than head; if longer, then forelegs enlarged for grasping	p.	72
30.	preyCostal cell with many crossveinsNEUROPTERA, suborder PLANIPENNIA,	p.	
2.1	Costal cell without series of crossveins————————————————————————————————————	•	71 32
3 1.	Hindwings smaller than forewings; tarsi 2- or 3-segmented		33
32.	Tarsi apparently 4-segmented; forebasitarsi unswollen; wings dehiscent (see also couplet 7)ISOPTERA, p).	65
33.	Tarsi 3-segmented, forebasitarsi swollenEMBIOPTERA, Cerci absent; wings remaining attached; antennae slender, with 13 or more segmentsPSOCOPTERA,		
	Cerci evident, although short, ending in bristle; wings shed eventually; antennae with 9 beadlike segments; seldom-encountered small insects		
34.	Body with more or less distinct head, thorax, and abdomen; with jointed legs and ability to move about		35
	Without distinctly separate body parts, or without legs, or not able to move about		78
35.	Parasites of warm-blooded animals		36
	Not parasites of warm-blooded animals		40
36.	Body strongly flattened sideways; mouth a sharp, downturned beak (jumping insects)SIPHONAPTERA,	•	
37.	Body flattened dorsoventrally or maggots of more or less cylindrical form Mouthparts with mandibles for chewing, directed forward; generally oval insects with more or less triangular head; parasites of birds and mammals		37
	MALLOPHAGA,	•	
38.	Mouthparts in form of beak for piercing and sucking Antennae inserted in pits, not visible from above (also maggot-shaped larvae without antennae)DIPTERA (a few families),		38 76
	Antennae present, although short, not in pits		39
39.	Beak not jointed; tarsi forming hook for grasping hairs of host; parasites remaining on hostANOPLURA,	p.	6 8
	Beak jointed; tarsi not hooked; parasites not remaining on host (bedbugs and related insects)HETEROPTERA,		
	Aquatic, usually breathing by gills; larval and some pupal forms————————————————————————————————————		41 49
41.	Mouth forming a strong, pointed, downcurved beakImmature HETEROPTERA,	p.	69
	Mouth with mandibles		42

42.	Mandibles extending straight forward, united with maxillae to form piercing jawsSome larval NEUROPTERA, p	. 72
	Mandibles moving sideways, forming biting jaws	43
43.	Living in case formed of sand, pebbles, leaves, twigs, etc.; usually with external tracheae serving as gillsLarvae of some TRICHOPTERA, p	. 72
	Not living in case	44
44.	Abdomen with lateral organs serving as gills (a few larval Trichoptera and	
	Coleoptera key out here also)	45
	Abdomen without external gills (some larval Trichoptera will key out here also)	46
45.	Abdomen with 2 or 3 long tail filamentsLarvae of EPHEMEROPTERA, p	. 61
	Abdomen with short end processes (larvae of some Trichoptera will run to this	
	point)Larvae of NEUROPTERA, suborder MEGALOPTERA, p	. 72
46.	Lower lip (labium) folded backward, extensible, and furnished with pair of	
	jawlike hooksLarvae of ODONATA, p Labium not so constructed	
47	Abdomen with nonjointed false legs (pseudopods) in pairs on several	47
47.	segmentsFew larvae of LEPIDOPTERA, p	72
	Abdomen without pseudopods	48
48	Thorax in 3 loosely united divisions; antennae and tail filaments long and	40
70.	slenderLarvae of PLECOPTERA, p	66
	Thoracic divisions without constrictions; antennae and tail filaments short	. 00
	(larvae of some aquatic Diptera and Trichoptera also run to this point)	
	Larvae of COLEOPTERA, p	.70
49.	Mouthparts retracted into head and hardly or not at all visible; underside of	
	abdomen with appendages; very delicate, small, or minute insects,	
	sometimes without antennae	50
	External mouthparts conspicuous; antennae always present; underside of	
	abdomen rarely with appendages	52
50.	Head pear shaped, without antennae; abdomen without long cerci, pincers, jumping apparatus, or basal ventral suckerPROTURA, p	61
	Head usually not pear shaped, antennae conspicuous; abdomen with long	
	cerci, pincers, or basal ventral sucker	51
51.	Abdomen consisting of 6 or fewer segments, with forked sucker at base below	
	and usually with conspicuous jumping apparatus near end, but without	
	conspicuous long cerci or pincersCOLLEMBOLA, p	. 61
	Abdomen with more than 8 evident segments and ending in long, many-jointed	
	cerci or strong pincers; eyes and ocelli lackingDIPLURA, p	
52.	Mouthparts with mandibles fitted for chewing	53
	Mouthparts in form of proboscis fitted for sucking	74
53.	Body usually covered with scales; abdomen with 3 prominent tail filaments	~ 4
	and at least 2 pairs of ventral appendages (styles)THYSANURA, p Body never covered with scales; abdomen never with 3 tail filaments nor	0. 61
		54
51	ventral styles Abdomen bearing pairs of false legs (pseudopods) beneath, not jointed, and	54
54.	differing from true legs on thorax, which is not distinctly separated from ab-	
	domen; body caterpillarlike; larval forms	55
	Underside of abdomen without legs or pseudopods	57
55.	Pseudopods 5 pairs or less, none on 1st, 2d, or 7th segments; pseudopods	٠.
	tipped with many tiny hooklets and rarely present on 2d and 7th segments	
	Larvae of most LEPIDOPTERA, p	. 73
	Pseudopods 6 to 10 pairs, not tipped with tiny hooks; 1 pair of pseudopods on	
	2d segment	56

57.	Antennae short; larval forms 7 Abdomen ending in strong pincerlike forceps; prothorax free	71 58 71
59.	Abdomen not ending in forceps	59
	Abdomen not strongly constricted at base, broadly joined to thorax————————————————————————————————————	60
62.	Cerci of single segment, prominent———————————————————————————————————	67
63.	Hindlegs fitted for jumping, femora enlarged; wing pads of immatures inverted, hindpads overlapping forepadsORTHOPTERA, p. 6	
64.	Prothorax much longer than mesothorax; front legs modified for grasping prey (raptorial)MANTODEA, p. 6	_
65.	Prothorax not greatly lengthened	65 70
67.	Cerci present; antennae usually with more than 15 segements	66 67 69 63 68
69.	Cerci short; ovipositor lacking; tarsi 4-segmented	65
70.	Front tarsi with 1st segment swollen, containing silk-spinning gland for	70
71	producing web in which insects live; cerci conspicuousEMBIOPTERA, p. 6 Front tarsi not so, not producing silk; cerci inconspicuousISOPTERA, p. 6 Body cylindrical, caterpillarlike	
	Body more or less depressed, not caterpillarlike	73
72.	Head with 6 ocelli on each side; antennae inserted in membranous area at base of mandiblesSome larvae of LEPIDOPTERA, p. Head with more than 6 ocelli on each side; 3d pair of legs distinctly larger than	73
72	1st pair	71
13.	Mandibles united with maximae to form sucking jaws	ne

74.	Body densely clothed with scales and hairs; proboscis, if present, coiled under headLEPIDOPTERA, p. 73
	Body bare, with scattered hairs or waxy coating 75
75.	Last tarsal segment bladderlike, without claws; mouth a triangular unsegmented beak; very small insectsTHYSANOPTERA, p. 68
	Tarsi not swollen at tip, with distinct claws 76
76.	Prothorax small, hidden when viewed from aboveDIPTERA, p. 76
	Prothorax evident when viewed from above 77
77.	Beak arising from front part of headHETEROPTERA, p. 69
	Beak arising from lower back part of headHOMOPTERA, p. 69
78.	Legless grubs or maggots, moving about by squirming Larvae of DIPTERA
	(if aquatic wrigglers, see larvae and pupae of mosquitoes); HYMENOPTERA; LEPI-
	DOPTERA; COLEOPTERA; SIPHONAPTERA; STREPSIPTERA (in body of wasps or
	bees with flattened head exposed)
	Forms legless or with a single claw 79
79.	Small forms with little resemblance to most insects, with filamentous mouthparts inserted in plant tissue; usually covered with waxy scale,
	powder, or cottony tuftsHOMOPTERA, p. 69
	Body unable to move or only able to bend from side to side, enclosed in tight
	skin, sometimes wholly covering body or sometimes with appendages free,
00	but rarely movable; sometimes enclosed in cocoon (pupae)
80.	Legs, wings, etc., more or less free from body; biting mouthparts visible 81
	Skin enclosing body holding appendages tightly against body; mouthparts
0.4	evident as proboscis, without mandibles 83
81.	Prothorax small, fused with mesothorax; sometimes enclosed in thin cocoon –
	Pupae of HYMENOPTERA, p. 78
00	Prothorax larger and not fused with mesothorax 82
82.	Wing cases with few or no veinsPupae of COLEOPTERA, p. 70
00	Wing cases with several branched veinsPupae of NEUROPTERA, p. 72
83.	Proboscis usually long, rarely absent; wing cases 4; often in cocoon
	Pupae of LEPIDOPTERA, p. 73
	Proboscis usually short; wing cases 2; rarely in cocoon, but often tightly
	enclosed in hardened last larval skinPupae of DIPTERA, p. 76

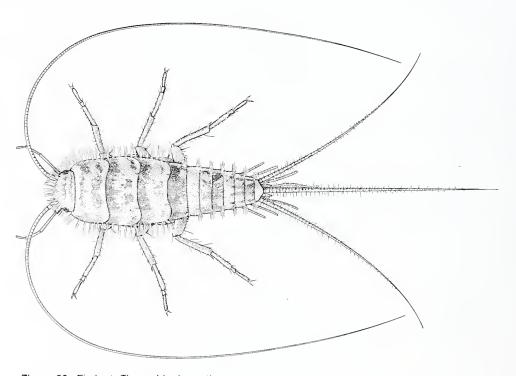


Figure 30. Firebrat, *Thermobia domestica* (Packard), order Thysanura.

Descriptions of Insect Orders

The following descriptions of the various orders are arranged according to the preceding synopsis (p. 54).

Subclass Apterygota

Of the four orders (1-4) included in the Apterygota, the Protura and Diplura (1 and 3) are of such slight importance to agriculture that they are not considered here.

All Apterygota should be collected and preserved similarly. They may be collected with a Berlese funnel, aspirator, or small brush dipped in alcohol. To collect the species of Collembola that are often found on the surface of water, use a small dipper (see ref. 97). Specimens should be killed and preserved in 80 percent alcohol. All Apterygota are delicate. To minimize risk of breakage, fill their vials full with fluid. Microscope slide preparations are necessary for critical work with all Apterygota.

Thysanura (Bristletails, Silverfish). Thysanura (fig. 30) are elongate, wingless insects with long antennae and with two or three taillike filaments at the end of the abdomen. The mouthparts are fitted for chewing and the metamorphosis is simple. The most common families are the Lepismatidae and Machilidae. Some species, such as Lepisma saccharinum Linnaeus and Thermobia domestica (Packard), are found in domestic situations feeding on bookbindings, curtains, wallpaper paste, paper, clothing, and similar articles. Most thysanurans occur outdoors under bark and stones, in grass and leaf litter, or in rotting wood or other debris.

Collembola (Springtails). Collembola (fig. 31) are tiny, wingless insects commonly called springtails. Most of them have a forked structure on the ventral side of the fourth abdominal segment that is held in place by a clasplike structure called a tenaculum on the third abdominal segment. When the insect jumps, it releases the forked structure with sufficient force against the surface of the ground so that it is propelled into the air. The mouthparts are concealed within the head, and metamorphosis is simple. Three commonly collected families are the Poduridae, Entomobryidae, and Sminthuridae. Springtails are found in rotting wood and leaf debris, on the surface of ponds and streams, in soil and fungi, under bark, and on vegetation. Some species are abundant on the surface of snow and have been given the name "snow fleas."

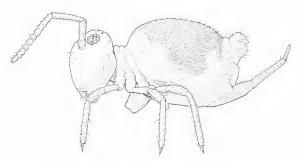


Figure 31. Springtail, *Bourletiella hortensis* (Fitch), order Collembola, family Sminthuridae.

Subclass Pterygota

Ephemeroptera (Mayflies). Mayflies (fig. 32, B) are soft-bodied, elongate insects with at least one pair of membranous wings and two or three long, slender tails. Adults do not feed, and their mouthparts are vestigial; those of the nymphal stages are fitted for chewing. The nymphs (fig. 32, A) are aquatic, with gills along the sides of the abdomen. Metamorphosis is simple. Mayflies possess a unique developmental stage, called a subimago, which is the initial winged form. This is not the adult stage: the subimago undergoes one more molt before becoming an adult. Three families are commonly collected—Ephemeridae, Heptageniidae, and Baetidae. Nymphs are found in a variety of aquatic habitats, from fast-flowing streams to the still waters of ponds, where some species burrow into the muck at the bottom. Adults are usually seen near water on vegetation and other objects, but at times they are attracted in large numbers to lights.

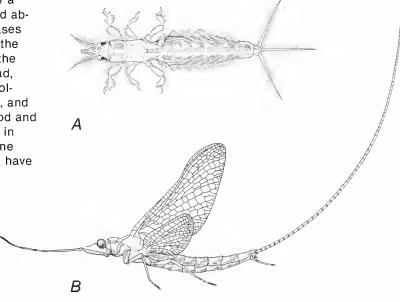


Figure 32. Typical mayflies, order Ephemeroptera: *A*, Immature nymph, *Stenonema canadense* (Walker); *B*, adult.

Mayfly nymphs can be collected with an aquatic net or can be handpicked from submerged rocks and vegetation. They should be killed and preserved in 80 percent alcohol or held and reared to the adult stage in aquariums or quart-sized or larger jars half filled with water. Although adult mayflies rarely live for more than a few days, nymphs often require an entire year to develop. Unless full-grown nymphs are collected, the collector should be prepared to keep the specimens in captivity for many months. Since some species need well-oxygenated water, a small electric pump to which air hoses can be attached may be necessary. Almost all mayfly nymphs are plant feeders, and an adequate supply of aquatic plants must be provided in the aquarium or rearing container. Some of the vegetation, a stick, or a rock should extend above the surface of the water to provide the subimago with something to which it can cling after emergence.

Subimagos can be preserved in alcohol as recommended for nymphs, but it is better to hold them in rearing cages until the final molt is completed. Both subimagos and adults can be captured with an aerial collecting net or by sweeping or beating the foliage near the water. Mayflies are extremely fragile, and particular care must be taken in removing them from a net so as not to damage their delicate appendages. Adults may be preserved in alcohol or pinned; however, pinned specimens usually shrivel badly and become brittle so that they are difficult to handle.

Odonata (Dragonflies, Damselflies). The order Odonata is divided into the suborders Anisoptera (dragonflies) and Zygoptera (damselflies). The former are generally large insects that usually keep their wings outstretched when at rest, whereas the latter are generally smaller, more delicate insects that usually fold their wings back over the abdomen when at rest. There are seven families of Anisoptera and three of Zygoptera. Dragonflies (fig. 33, A) and damselflies (fig. 33, B) are comparatively large and have two pairs of many-veined wings. The hindpair is as large as or larger than the front pair. The antennae are short and bristlelike, the abdomen is long and slender, the mouthparts are fitted for chewing, and the metamorphosis is simple. The nymphs are aquatic, possess gills (internal in dragonflies), and have the labium modified into a scooplike structure that can be rapidly extended forward to grasp prey.

The nymphs may be found clinging to aquatic vegetation or in the muck at the bottom of streams or ponds. Specimens are easily reared on a diet of young tadpoles, aquatic insects, or other small aquatic animals. A long stick placed in the rearing tank allows mature nymphs to crawl out of the water for the final molt to the adult stage. Nymphs should be preserved in alcohol; large ones should be killed in boiling water, then transferred to alcohol.

Dragonflies, especially the larger ones, are fast fliers and are most easily caught when resting. Some have

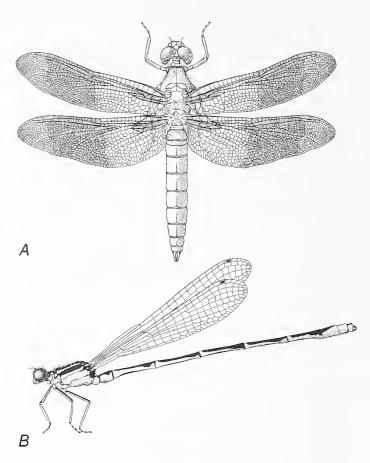


Figure 33. *A*, Pond dragonfly, *Plathemis lydia* (Drury), order Odonata, family Libellulidae; *B*, damselfly, *Enallagma exsulans* Hagen, order Odonata, family Coenagrionidae.

favorite resting places and fly regular routes, so collectors with sufficient patience to wait quietly may be rewarded by having the insect come to them. Any strange and sudden movement, as with a collecting net, may cause the dragonfly to dart away. Try to swing the net from behind and a little below the insect, where it will be less apt to see the movement until it is too late. Captured specimens may be transferred directly for half a day to acetone, which preserves the colors well, placed in a cyanide jar, where some colors may be lost, or held alive for a day or two without food. The purpose of the last is to eliminate the contents of the intestinal tract before the insect is killed. Decomposing food left in the body of a dry specimen may affect some colors. If held alive, adults should be confined to a small space and preferably kept in the dark so they will not beat and damage their wings. Adults obtained by rearing should be held alive for a time to allow the colors to develop fully, but newly emerged adults should be kept in cages large enough to permit the wings to expand fully. For permanent collections, pin adults with the wings spread or place them in clear plastic envelopes with the wings folded above the dorsum. With pinned specimens, brace pins may be added on either side of the abdomen for support. This is especially important if the specimens are to be mailed.

Orthoptera (Crickets, Grasshoppers, Katydids). Members of the order Orthoptera have the hindlegs enlarged and adapted for jumping. The front wings are thickened, the cerci are unsegmented, the mouthparts are fitted for chewing, and metamorphosis is simple. The larvae (nymphs) resemble the adults. There are 11 families in this order.

Some crickets (fig. 34, A) are found in domestic situations, but most orthopterans inhabit vegetation such as grass and shrubs on which they feed. Although they usually move by walking or jumping, most adult grasshoppers can fly, often exposing brightly colored hindwings. To capture the adults in a net, watch where they land and bring the net down quickly over the spot. If the tip of the bag is held upright, the grasshopper usually can be coaxed up into the bag. Both adults and nymphs can be captured by sweeping vegetation. Flowerpot cages are ideal for rearing grasshoppers.

Most species of katydids (fig. 34, *B*) are nocturnal. Many are attracted to light and usually can be caught easily by hand. Crickets also are active at night and may be attracted to baits such as molasses or oatmeal, which has been prepared as a paste and spread in a thin layer. The paste is made by mixing dry oatmeal with a little sugar, dry milk, and water. This same dry paste can be used to feed crickets in captivity, although the diet should be supplemented occasionally with bits of fruit and lettuce.

Both adults and larvae of Orthoptera can be killed and preserved in alcohol; for a permanent collection, pinned adults are preferable. For demonstrations or exhibits, winged forms may be mounted with one or both pairs of wings spread, but for most collections the specimens should be mounted with legs, antennae, and wings folded close to the body to conserve space.

Blattodea (Cockroaches) and Mantodea (Mantids). In both the Blattodea and Mantodea, the hindlegs are similar to the middle ones and are adapted for walking. The antennae have more than 30 segments, the cerci are many-segmented, the mouthparts are fitted for chewing, and the metamorphosis is simple.

Cockroaches (fig. 35) are general feeders, attacking both plant and animal material. There are over 50 species of cockroaches in North America, of which only 4 are common indoors. These domestic cockroaches are active at night and usually avoid light. Some outdoor species, on the other hand, may be attracted to light at night, and they may also be attracted to pitfall traps baited with molasses. This attraction for sweets sometimes leads cockroaches to invade abandoned beehives to feed on the remaining honey. During the day, most cockroaches hide under the loose bark of trees, beneath rotting logs, or in similar fairly moist habitats. They can be reared with little difficulty, and many species are available from biological supply houses.

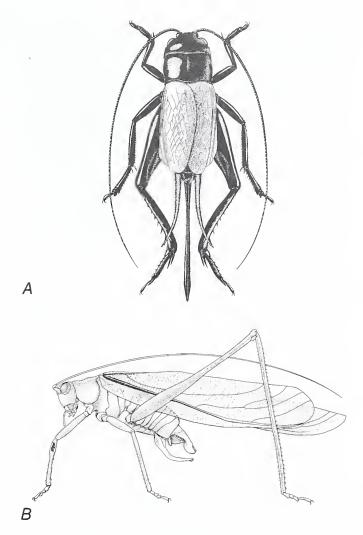


Figure 34. *A*, Cricket, order Orthoptera, family Gryllidae; *B*, katydid, order Orthoptera, family Tettigoniidae.

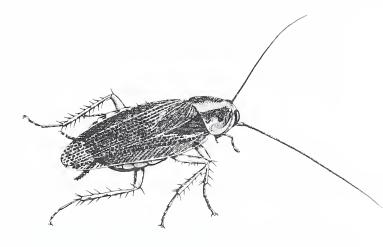


Figure 35. German cockroach, Blattella germanica (L.), order Blattodea, family Blattellidae.

Mantids (fig. 36) are generally found on vegetation, where they prey on other insects. During the autumn and winter, mantid egg cases can be found attached to low shrubs or grass stems. If collected, it is suggested that the egg cases be kept outdoors or in an unheated garage or porch until spring to prevent the eggs from hatching prematurely unless a quantity of living insects such as Drosophila flies and aphids is available to feed the young nymphs. Adult mantids normally die in the fall soon after the eggs are laid, but sometimes specimens can be kept alive for several weeks longer indoors by careful feeding in rearing cages. Mantids are voracious feeders, and their egg cases are sold by many garden suppliers, who advertise that the mantids will control numerous garden pests. The nymphs and adults consume large numbers of insects, but only those readily visible on plant foliage. Soil-dwelling or stalk-boring insects are safe from attack. Despite their formidable appearance, mantids can be safely captured by hand. Specimens may also be collected from vegetation with a sweep net.

Adults and nymphs of cockroaches and mantids may be preserved in alcohol, but for a permanent collection, adults should be pinned.

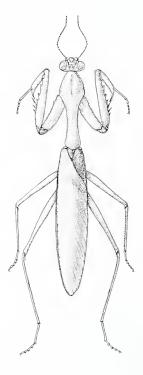


Figure 36. Carolina mantid, *Stagmomantis carolina* (L.), order Mantodea, family Mantidae.

Phasmatodea (Walkingsticks). Phasmatids or walkingsticks are elongate, slow-moving insects with all legs similar in appearance and adapted for walking. Most species are wingless. The cerci are short and unsegmented, the mouthparts are fitted for chewing, and metamorphosis is simple. The nymphs resemble the adults.

Walkingsticks are found on trees and shrubs, on which they feed. Because of their slow movements, resemblance to twigs, and protective coloration (usually green or brown), they are difficult to see. Sweeping or beating trees and shrubs may yield some individuals. In spring or early summer, newly hatched nymphs may be found, which in the most common species, *Diapheromera femorata* (Say), are only about 0.5 cm long. The nymphs mature in about 6 weeks, attaining a length of 8-10 cm. Their slow-moving, plant-feeding habits enable them to adapt readily to the environment of a rearing cage. Although normally phasmatids are leaf feeding, in captivity the newly molted nymphs have been observed eating the cast skin.

Both adults and immatures may be collected by hand and dropped into alcohol or a killing jar. If specimens are pinned, additional brace pins may be necessary to support the abdomen.

Grylloblattodea (Rock Crawlers). These are rare insects found along the edges of glaciers. They are of no economic importance.

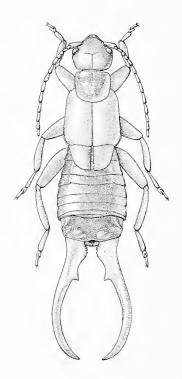


Figure 37. Male European earwig, *Forficula auricularia* L., order Dermaptera, family Forficulidae.

Dermaptera (Earwigs). Members of the order Dermaptera (fig. 37) can be recognized by their distinct, pincerlike cerci at the apex of the abdomen. The adults usually have two pairs of wings. The front pair is leathery and shorter than the abdomen; the hindpair is membranous and folded under the front wings when the insect is at rest. The mouthparts are fitted for

chewing, and metamorphosis is simple. The eggs are laid in a burrow in the ground, and the female cares for them until they hatch, a situation rarely seen in the insect world. The nymphs resemble the adults. In North America, the families of earwigs are the Forficulidae, Chelisochidae, Labiduridae, and Labiidae.

Contrary to the old superstition from which the common name is derived, earwigs are not known to crawl into the ears of sleeping people. However, they are nocturnal insects. During the day, they usually hide in cracks and crevices, under the bark of trees, in rubbish piles, or beneath stones, leaves, and other debris in or on the ground. One species, Anisolabis maritima (Géné), is commonly collected under stones or driftwood along the Atlantic and Pacific coasts. Earwigs feed as scavengers on decaying organic matter, but some species may also feed on living tissues of flowers, ripening fruits and vegetables, and occasionally on aphids and other small insects. Despite their almost omnivorous feeding habits, or perhaps because of them, earwigs are not strongly attracted to bait traps. It is usually necessary to search by day in likely habitats and collect the specimens by hand. Adults and nymphs may be killed and preserved in alcohol, but for a permanent collection the adults should be pinned.

developed social system, and both winged and wingless adults may occur in a colony. The winged form is the reproductive caste, composed of fertile adults with compound eyes and comparatively dark coloration. After the dispersal flight from the parent colony, the wings are usually shed, leaving only short stubs that are easily visible under low magnification. Wingless forms are sterile adults of either the worker or soldier castes. Workers provide food for the colony, construct new tunnels and chambers, and care for the egg-laving queen, whose abdomen is so swollen that she is immobile. Soldiers defend the colony against attack. Their heads are greatly enlarged, usually with strong biting jaws, but in some species the jaws are supplemented with a beak through which a fluid may be ejected to repel enemies. The principal families in the order are the Hodotermitidae, Kalotermitidae, Rhinotermitidae, and Termitidae.

Based on habitat, termites may be divided into two main types, nonsubterranean and subterranean. The nonsubterranean termites include species that live in dead trees, wooden buildings, or furniture not in contact with the soil. Subterranean termites live in the soil, sometimes creating mounds protruding a few meters above the surface. Colonies of subterranean termites

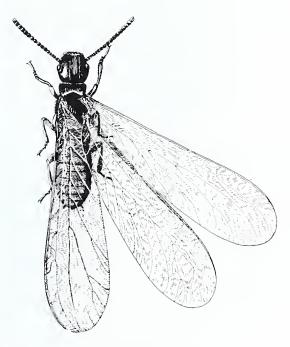


Figure 38. Eastern subterranean termite, *Reticulitermes flavipes* (Kollar), order Isoptera, family Rhinotermitidae.

Isoptera (Termites). Termites (fig. 38) are small, soft-bodied insects with simple metamorphosis and mouth-parts fitted for chewing. Although often called "white ants" or "flying ants," termites may be distinguished from true ants (fig. 39) by having a broad waist, beaded antennae, and, on winged forms, having both pairs of wings the same size and shape. Termites have a highly

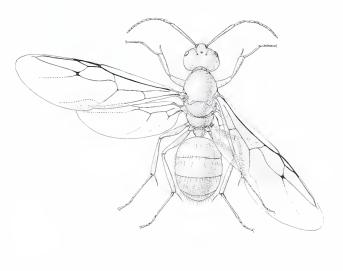


Figure 39. Typical winged-caste ant, *Camponotus castaneus* (Latreille), order Hymenoptera, family Formicidae.

may be located by digging into these mounds, by prying apart rotting stumps, or by turning over rotting logs and searching in the soil beneath. Colonies of nonsubterranean termites are more difficult to find, since often there is no external evidence of an infestation. Careful examination may show the entrance holes made by the reproductive adults as they entered the wood, but these

holes usually are sealed with cementlike plugs secreted by the termites. If the termites are working close to the outer edge of the wood, surface blisters in the paint or a flaking away of the surface of unpainted wood may be a clue to the presence of a colony.

Using an aspirator, collect representatives of each of the various castes if possible, as identification often cannot be made from the workers alone. Do not overlook insects of other orders that often live in the colony and that may mimic termites in color and general appearance. All castes of termites should be killed and preserved in alcohol.

Embioptera (Webspinners). Embioptera are small insects, mostly 4-7 mm long, that live in silken galleries, which they spin in soil, on or under bark, among mosses and lichens, or sometimes in stored products. A few species are found in the Southern United States and in California, but they are of no economic importance. Embiopterans may be treated as termites for a collection.

Plecoptera (Stoneflies). Adult stoneflies (fig. 40) are soft-bodied insects with two pairs of wings except in a few species in which the wings of the males may be lacking or greatly reduced. The legs and antennae are long, and usually there are two long, taillike appendages. Sometimes, as in the family Nemouridae, the "tails" are short and modified into accessory reproductive organs. The mouthparts are fitted for chewing but may be poorly developed in species that do not feed as adults. Stonefly larvae are aquatic, with long antennae and cerci. Branched gills, if present, are located on the thorax at the base of the legs. The larvae resemble those of mayflies but differ in the location of the gills and in having two tarsal claws instead of one. The mouthparts of the nymphs are fitted for chewing, and metamorphosis is simple. In North America, the order has nine families. Members of the family Perlidae are most common.

Stonefly larvae are found in running water, under stones, or clinging to submerged piles of drifted leaves and other debris. Specimens can be collected with an aquatic net, a dipper, or by hand, and dropped into vials of alcohol or held for rearing. One difficulty in rearing Plecoptera is in determining the food preferences of the nymphs collected. The species most commonly collected are plant feeders, but some species are carnivorous. To reduce the time needed for rearing, many collectors schedule their collecting to coincide with the adult emergence season of a particular species of Plecoptera. The adults almost always emerge at night or early in the morning. By visiting streams at night, one can collect mature larvae as they crawl out of the water just prior to their final molt, and they may be held in temporary cages until the adults emerge.

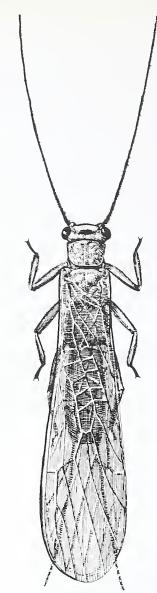


Figure 40. Common stonefly, Neoperla clymene (Newman), order Plecoptera, family Perlidae.

Most insects hibernate or become inactive in cold weather, but adults of many species of Plecoptera emerge during the winter, even in the northern parts of our country. Their dark forms against the snow make them conspicuous and easy to collect. Species that mature in spring or summer often fly to lights at night. During the day, adults are usually found resting on vegetation or other objects along streambanks and can be collected by sweeping or beating the foliage. Most stoneflies are poor fliers but agile runners. Because they generally are slow to take flight, specimens often can be picked up by hand or brushed off foliage with a flick of the finger directly into a vial of alcohol.

Both adults and nymphs can be preserved in alcohol. If pinned, adults may become shriveled, but many collectors prefer this method of preservation. Wings, genitalia, and other structures critical for identification may have to be dissected and mounted on slides or placed in microvials.

Psocoptera (Psocids, Booklice). Members of the order Psocoptera (fig. 41) are small, soft-bodied insects somewhat resembling psyllids or aphids (fig. 42) but differing from them in having chewing mouthparts. The wings, if present, are membranous with characteristic curved, intermeshing veins and few if any crossveins. When the insect is at rest, the wings usually are held rooflike over the back. Metamorphosis is simple. Eleven families are found in North America.

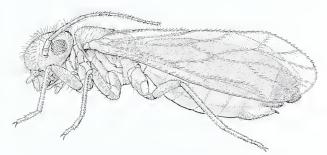


Figure 41. Winged psocid (barklouse), *Ectopsocus pumilis* (Banks), order Psocoptera, family Pseudocoeciliidae.

Some usually wingless species found on old books or papers have received the common name "booklice." In damp locations in houses and granaries, booklice may attain tremendous numbers. Most species of Psocoptera occur outdoors on the trunks and leaves of trees and shrubs, on lichen-covered stones, and on fences. Psocids feed on mold, lichen, pollen, cereals, and starchy materials. Unfortunately, they also feed on dead insects and can cause considerable damage in an insect collection unless preventive measures are taken (see p. 43).

Specimens can be collected by sweeping or beating vegetation, by drying leaf debris in a Berlese funnel, or by picking specimens from tree trunks or rocks with an aspirator or fine brush moistened in alcohol. Both adults and nymphs should be preserved in alcohol.

Zoraptera (Zorapterans). Zorapterans are small insects not over 3 mm long, usually found under planks, in piles of old sawdust, in rotting logs, and under bark. Only two species are found in North America; they occur from Pennsylvania and lowa to Texas and eastward. They are of no economic importance. They may be treated as termites for a collection.

Mallophaga (Chewing Lice). Chewing lice (fig. 43) are small, wingless, flattened external parasites of birds and mammals, although mostly of birds. The mouthparts are fitted for chewing, and metamorphosis is simple. The immatures resemble the adults, and all stages occur on the host; in fact, lice are rarely found away from the host. Of the six families commonly found in North America, the Philopteridae on poultry and the Trichodectidae on such mammals as cattle, horses, and dogs are perhaps the most often collected. No species of Mallophaga are known to attack man.

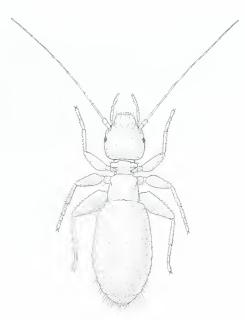


Figure 42. Wingless psocid (booklouse), *Liposcelis divinitorius* (Mueller), order Psocoptera, family Liposcelidae.

Chewing lice are collected from the living or dead host. All parts of the host should be examined for the lice because different species of Mallophaga may be found on different parts of the host. In labeling the specimens, always include as part of the collection data the part of the host's body from which the parasites were taken. Use forceps or a fine brush moistened in alcohol to remove the lice from the feathers or hairs of the host. Drop the specimens into small vials of alcohol, using a separate vial for each species and for specimens from different hosts. For permanent collections, the lice should be stained if necessary and mounted on microscope slides.

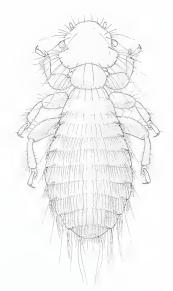


Figure 43. Chewing louse, *Meromenopon meropis* Clay and Meinertzhagen, order Mallophaga, family Menoponidae.

Anoplura (Sucking Lice). Sucking lice (fig. 44) are similar to chewing lice in that they are small, wingless, and flattened, but instead of chewing, they suck blood and are parasites of mammals only. Metamorphosis is simple, with the larvae resembling the adults, and all stages are found on the host. This order contains species that attack humans. The crab louse, Pthirus pubis (Linnaeus), the head louse, Pediculus humanus capitis De Geer, and the body louse, P. humanus humanus Linnaeus, are the species usually found on humans. The body louse is an important vector of typhus and other diseases. In the United States, three families in the order are found—the Pediculidae (to which the three species mentioned here belong), the Echinophthiridae (on marine mammals), and the Haematopinidae (on horses, cattle, sheep, hogs, and other animals).

Anoplura must be collected from the host. The methods of collection and preservation are the same as for Mallophaga.

Thysanoptera (Thrips). Thrips (fig. 45) are minute insects that may be winged or wingless. The winged species have two pairs of wings with few or no veins and are fringed with long hairs. The piercing-sucking mouthparts are enclosed in a conical, asymmetrical structure on the ventral side of the head. Metamorphosis is intermediate between simple and complete. The immature stages resemble the adults as in simple metamorphosis. Because the fourth instar is inactive, does not feed, and may have external wing pads, it is called a pupa. The earlier instars are called larvae and prepupae as in complete metamorphosis. The prepupal and pupal stages may be enclosed in a cocoon. Five families of thrips are found in North America.

Most thrips feed on flowers, pollen, leaves, buds, twigs, and other parts of plants; some thrips are predaceous on other small arthropods; and a few species feed on fungus spores. Specimens can be collected by sweeping vegetation or by shaking or beating shrubbery over a white or light-colored pan. Another method is to collect infested plant parts, usually obvious because of the curled leaves or deformed buds, and place the vegetation in a paper bag. These samples can be examined later and the thrips carefully picked off with an aspirator or a fine brush dipped in alcohol, or the sample can be run through a Berlese funnel. Collect winged and wingless forms if both are present. Thrips can be collected in 60-70 percent alcohol, but AGA (see p. 94) is recommended because it leaves the appendages well extended and makes the specimens easier to mount on slides.

For critical study and for permanent collections, thrips should be mounted on slides. Specimens need not be stained before mounting, but some dark-colored and spore-feeding thrips must be treated with cold 10 percent sodium hydroxide to lighten the color and remove the body contents. Do *not* use potassium hydroxide, which destroys the wings before the harder parts of the body are sufficiently treated. In some species, the

wings are held close to the body by paired setae on either side of the abdomen when the insect is at rest. To spread the wings, insert a needle between the wings and the body at the point where the thorax and abdomen join, and gently tease the wings out to a horizontal position. The antennae also should be extended because the number of antennal segments and position of sensoria on the segments often are critical for identification to species or even to family.

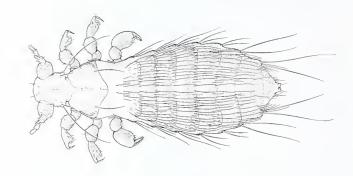


Figure 44. Sucking louse, *Polyplax stephensi* (Christopher and Newstead), order Anoplura, family Haematopinidae.

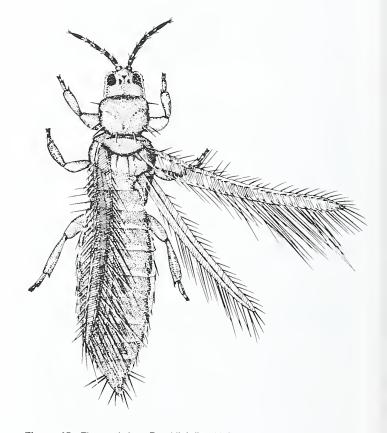


Figure 45. Flower thrips, *Frankliniella tritici* (Fitch), order Thysanoptera, family Thripidae.

Hemipterous Orders ("True" Bugs, Aphids, Cicadas, Leafhoppers, and Others). The order formerly known as Hemiptera is divided here into two orders, the Heteroptera (fig. 46) and the Homoptera (fig. 47).

In both orders, two pairs of wings are usually present and the mouthparts are fitted for piercing and sucking. Metamorphosis is simple, although members of the homopterous family Aleyrodidae pass through a pupal stage, males of scale insects pass through a prepupal, pupal, or both stages, and the life stages of the Phylloxeridae are even more complex. In most species

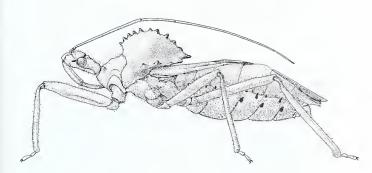


Figure 46. Masked hunter, *Arilus cristatus* (L.), order Heteroptera, family Reduviidae.

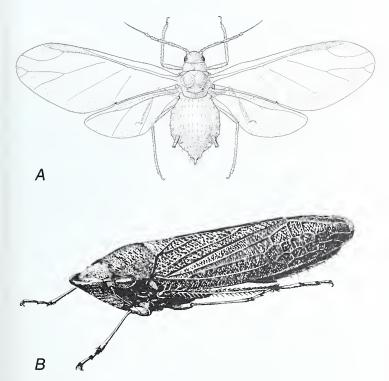


Figure 47. *A*, Winged aphid, *Rhopalosiphum padi* (L.), order Homoptera, family Aphididae; *B*, leafhopper, *Draeculacephala minerva* Ball, order Homoptera, family Cicadellidae.

the immatures resemble the adults. Members of the Heteroptera (fig. 46) have the basal part of the front wings thickened and leathery and the apical part membranous. The hindwings are entirely membranous and are covered by the front wings except in flight. When at rest, the wings are held flat over the back with the apical parts of the front wings overlapping. The mouthparts, in the form of a beak, arise from the front part of the head.

Although the term "bug" is used loosely to refer to all insects, in the strict sense only species of these two orders are true bugs. Members of the Homoptera are similar to those of the Heteroptera except the sucking mouthparts arise from under the back of the head and the front wings are uniform in texture, either entirely membranous as in the aphids (fig. 47, A) or of a slightly thicker, almost leathery texture as in the leafhoppers (fig. 47, B). The hindwings, absent in male scale insects, always are entirely membranous. When at rest, members of this order hold the wings folded rooflike over the back.

Members of the order Heteroptera are very diverse in appearance and habits. Although most species are terrestrial, many of the true bugs live in or on the water. Many species are predaceous on other insects, a few suck the blood of humans and other vertebrate animals, but most, including all Homoptera, are plant feeders. Collecting methods for Heteroptera depend largely on the type of habitat. Most species are collected readily in nets, aquatic nets for species in or on the water and sweep nets for most plant-feeding and predaceous species. Many specimens of both orders, particularly during the mating season, are attracted to lights, and winged aphids are often attracted to yellow pan traps. However, because swept or trapped specimens lack associated host data, it is better to collect specimens directly from plants whenever possible. Collect both winged and wingless forms if both are present, and note the color of the living specimens; this color may be lost in the preservative or when the specimens are cleared and mounted on slides. Sifting or using a Berlese funnel on leaf litter or on soil around plant roots usually will yield some Heteroptera. Some species are found in association with ants and may mimic the ants in appearance. Species of Cimicidae (Heteroptera, bedbugs), which are ectoparasites of birds, bats, and other animals including humans, usually feed at night on their host and hide in cracks and crevices by day. Because cimicids are virtually wingless, they can be collected easily with small forceps.

With the possible exception of some fragile plant bugs of the family Miridae, both immature and adult Heteroptera can be killed and preserved in alcohol. Mirids and most Heteroptera can be collected satisfactorily in killing jars, but never put large specimens in the same jar with mirids or other delicate bugs. For permanent preservation, adults should be pinned. Specimens too small or slender for direct pinning should be glued to points on double mounts.

Most specimens of Homoptera can be collected and preserved, at least temporarily, in alcohol. Exceptions are diaspidid scales and whitefly pupae, which should be collected dry on the host plant and placed between pieces of absorbent paper. Do not use plastic bags. For critical study, aphids, whiteflies, and scale insects should be stained or bleached as needed, then mounted on slides. Adult cicadas, leafhoppers, and other relatively hard-bodied Homoptera can be collected in killing jars, then pinned. Specimens smaller than 10 mm should be glued carefully to points. When collecting gall-forming psyllids, include a sample of the gall. If securely anchored with additional pins, the gall can be kept in the box with the pinned adults.

Aphids and many other plant-feeding Homoptera can be maintained easily in flowerpot cages. In addition to allowing all stages of the insect to be observed and collected, the living specimens provide a ready source of food for young preying mantids and other predaceous insects in captivity.

Coleoptera (Beetles). Beetles (fig. 48) usually have two pairs of wings. The front wings are thickened, usually hard or at least leathery, lack veins but often are sculptured with rows of hollows or pits, and usually meet in a straight line down the middle of the back. The hindwings are membranous and are folded under the front wings when the insect is at rest. In some beetles, the wings are greatly reduced or absent. The mouthparts are fitted for chewing, and metamorphosis is complete. Several different types of larval forms occur within the order, none resembling the adults. This order contains about 40 percent of the species in the class Insecta and is the largest of the insect orders. More than 25,000 species occur in the United States alone. There are approximately 120 families, which are grouped into 3 suborders.

Beetles have invaded almost every conceivable habitat. There are both aquatic and terrestrial forms. There are plant or fungi feeders, predators, parasites, inquilines (welcome or unwelcome guests in nests of other kinds of insects), and scavengers. Some of the last, such as dermestid beetles, feed on dead insects and can be a problem in insect collections unless protective measures are taken. Plant-feeding beetles may be found in every part of a plant, from flowers to the roots. Some are external feeders; others mine leaves or bore into the stalk.

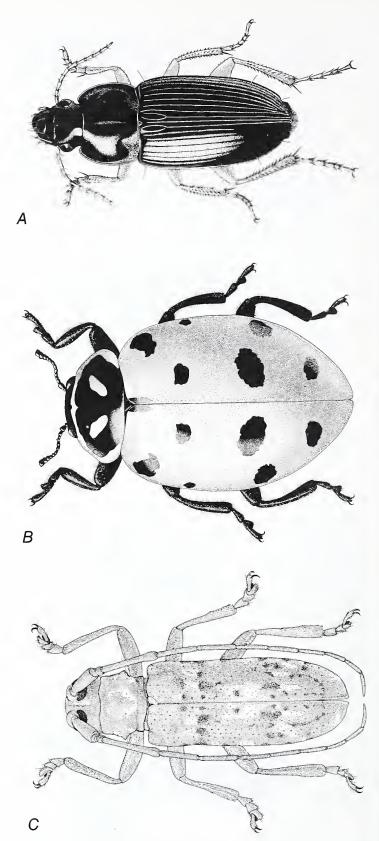


Figure 48. A, Ground beetle, Harpalus pennsylvanicus De Geer, order Coleoptera, family Carabidae; B, ladybird beetle, Hippodamia convergens Guérin-Meneville, order Coleoptera, family Coccinellidae; C, long-horned beetle, Monochamus titillator (Fab.), order Coleoptera, family Cerambycidae.

Collecting methods for beetles are almost as diverse as their habits. Pitfall and other bait traps using fermenting fruit or decaying meat will attract silphids, staphylinids, nitidulids, and other scavengers. Scarabs, cerambycids, and many other beetles are attracted to light traps or light sheets. A sifter or Berlese funnel can be used effectively to collect soil-dwelling beetles or beetles found in moss and leaf litter. Species living in ponds and streams can be captured by hand or with an aquatic net. Do not overlook beetles living along the shore in sand and under seaweed or other debris. Sweeping or beating vegetation will gather in many leaf-feeding and some predaceous beetles. A tool for getting under bark or into wood is necessary for collecting species that bore into logs or plant stems.

Immature beetles (larvae) should be killed by placing them in boiling water for 1-5 minutes, depending on the size of the specimens. They can then be preserved in alcohol. Adult beetles may be killed and preserved in alcohol, but it is recommended that they be killed in alcohol or in a killing jar and then mounted on pins. Specimens of most beetles should have the genitalia removed and prepared (see p. 37) after removal from alcohol before the specimen is pinned. Specimens smaller than 5 mm should be glued to points, using care not to conceal ventral characters. It is generally inadvisable to use minutens or to spread the wings of beetles. Very tiny or very flat, cleared beetles may be mounted on slides.

Many Coleoptera are easily reared; this is often desirable since it allows association of adults and immature stages. Mealworms and other larvae that infest stored grain and flour are particularly easy to rear and can be used to feed predaceous insects in captivity.

Strepsiptera (Twisted-Winged Parasites). Strepsipterans are very small, usually parasitic insects that are sometimes grouped with the Coleoptera. They are hypermetamorphic, that is, the first larval stage (triungulin) is very active and bears appendages, but after finding a host, such as various species of Orthoptera, Heteroptera, Hymenoptera, or Thysanoptera, the larva molts into a legless, wormlike form and pupates within the last skin of the host. The adult male is winged and leaves the host and flies about, whereas the female remains in the host with its body protruding from between the abdominal segments of the host, and after producing large numbers of the tiny triungulins, it dies. In males, the forewings are modified into small clublike or scalelike organs, and the hindwings are broad and delicate with few veins.

Strepsiptera males may sometimes be found on flowers, apparently searching for females in their hosts. However, the best way to collect strepsipterans is to capture and rear parasitized hosts, which may be recognized by the little saclike females protruding from the often distorted abdomens of their hosts. Specimens should be preserved in alcohol or mounted on microscope slides, but males may be mounted on minutens.

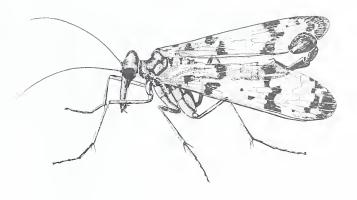


Figure 49. Scorpionfly, *Panorpa rufescens* Ramber, order Mecoptera, family Panorpidae.

Mecoptera (Scorpionflies). Mecoptera (fig. 49) have received the common name of scorpionflies because the upturned genitalia of some males resemble the tail of a scorpion. The adults usually have two pairs of long, narrow, membranous wings with many veins, although in some species the wings are reduced or absent. The mouthparts are fitted for chewing and are prolonged ventrally into a beaklike structure. Metamorphosis is complete. Larvae of scorpionflies resemble the larvae of sawflies and some Lepidoptera but differ from the latter by the absence of crochets (tiny hooks) on the prolegs and from the former by having seven or more ocelli. Families found in North America are the Boreidae, Bittacidae, Panorpidae, Panorpodidae, and Meropeidae.

Scorpionfly adults and larvae feed primarily on living and dead insects, but some adults are attracted to nectar and fermenting fruit, and some species apparently feed on moss. Larvae of most scorpionflies are found in the soil or in leaf litter on the ground and can be collected in a sifter. Most adults are found in heavily wooded areas and can be collected by sweeping or beating the vegetation. Adults of the so-called snow scorpionflies of the family Boreidae emerge in winter and often can be picked off the surface of snow with forceps. On rare occasions, some Mecoptera are collected in light traps, but as most species are active only during the day, flight traps such as the Malaise trap are usually more effective.

Larvae and adults can be killed and preserved in alcohol, but it is preferable to mount adults on pins or points. The wings of the adults may be spread if desired, but it is customary to leave them folded.

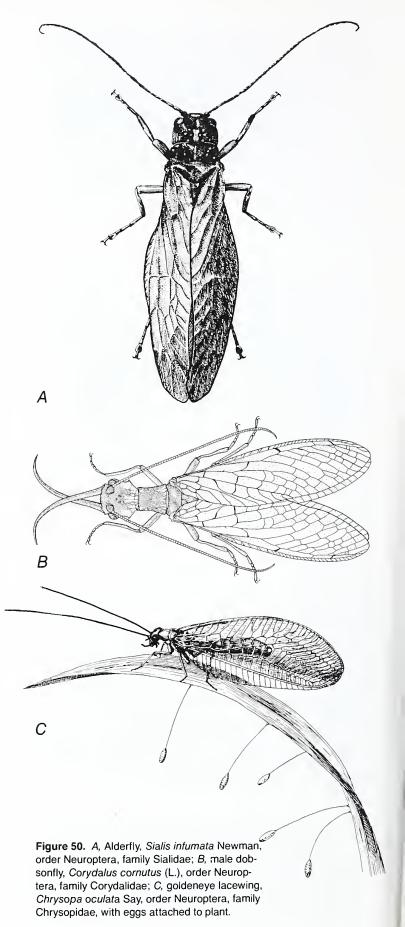
Neuroptera (Lacewings, Antlions, Snakeflies, Dobsonflies, Alderflies, and Others). Neuroptera (fig. 50) have two pairs of similarly shaped membranous wings, usually with a network of many crossveins and branches of longitudinal veins, particularly at their outer margins. When the insect is at rest, the wings are held rooflike over the back. The mouthparts are fitted for chewing. Metamorphosis is complete; the immatures do not resemble the adults. Classification of this group varies with different authors, but there are approximately 3 suborders and 14 families. Neuropterans such as dobsonflies and fishflies (family Corydalidae), alderflies (family Sialidae), brown lacewings (family Hemerobiidae), green lacewings (family Chrysopidae), and antlions (family Myrmeleontidae) are frequently found.

Most Neuroptera are predaceous both as adults and larvae, and many will bite if handled incautiously when collected. The larvae of several members of the order are aquatic, usually concealed under stones in streams, and can be collected by hand, in an aquatic net, or with a dipper. Adult aquatic forms usually remain on vegetation near water. They are relatively poor fliers and generally can be collected directly into killing jars. Most immatures and adults of terrestrial forms are found on vegetation and can be collected in a sweep net. Immature myrmeleontids, called antlions, are found partially buried at the bottom of small pits that they dig in sand or dust to trap ants or other unwary insects. The rarely collected mantispid larvae are parasitic on spider eggs. Many adults of both terrestrial and aquatic forms are attracted to lights.

Adult Neuroptera may be preserved in alcohol, but it is preferable to mount the specimens on pins or points. The wings may be spread or left folded. Immature specimens of medium to small size should be killed and preserved in alcohol; large specimens should be killed in nearly boiling water, then transferred to alcohol.

Trichoptera (Caddisflies). Caddisflies (fig. 51) are softbodied insects with long, slender antennae and two pairs of membranous wings, which are clothed with hairs and held rooflike over the back when the insect is at rest. The larval mouthparts are fitted for chewing but are modified in the adult for feeding on liquids. Metamorphosis is complete. About 20 families are in the order.

Trichopterous larvae, which are aquatic, have a pair of hooklike appendages at the end of the abdomen, and many live in characteristic cases constructed of pebbles, sand grains, twigs, or other materials found in ponds and streams. The abdominal hooks are used to drag the case about as the larva feeds. When the larva is ready to pupate, it attaches the case to a rock or other fixed object in the water. Case-making larvae are mostly detritus or plant feeders. Some caddisfly larvae



do not make cases but instead spin silken webs, which are used to capture food drifting in the stream. A few species build neither cases nor webs but are free living and predaceous. When collecting immature Trichoptera, preserve the case in alcohol along with the larva, pupa, or both.

Adult caddisflies are weak, usually crepuscular (dawn or dusk) fliers. They are generally found during the day resting on vegetation, bridges, or other objects near ponds and streams. Specimens can be collected with a sweep net or captured directly with a killing jar. Large numbers of adults are often attracted to lights, particularly if the lights are close to water. Adults killed and preserved in alcohol are satisfactory, but specimens mounted on pins are preferred. Adults too small for direct pinning should be mounted on minutens.

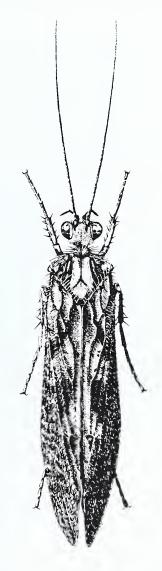


Figure 51. Caddisfly, *Phryganea vestita* (Walker), order Trichoptera, family Phryganeidae.

Lepidoptera (Butterflies, Skippers, Moths). Adult Lepidoptera (fig. 52) have two pairs of wings (rarely reduced or absent), which usually are covered with flattened hairs and scales. The mouthparts are fitted for sucking and are commonly in the form of a long, coiled haustellum (tongue), but this may be reduced or absent in nonfeeding adults. Metamorphosis is complete. The larvae (fig. 53) are commonly called caterpillars. They usually have a well-differentiated head with chewing mouthparts and a cylindrical, 13-segmented body. There are 3 pairs of thoracic legs and up to 5 pairs of prolegs, with apical crochets on abdominal segments 3-6 and 10. Part of the order is generally divided into two major superfamilies. One superfamily is represented by the butterflies (fig. 52, A) and the skippers (fig. 54), which are day fliers and have knobbed or clubbed antennae. The second superfamily includes the moths (fig. 52, B), which fly mostly at night and have threadlike or featherlike antennae. This is the second or third largest order of insects, with 2 suborders, about 78 families, and more than 150,000 species.

Most adults fly actively. Butterflies in particular often are collected on the wing or netted as they come to rest on flowers, leaves, or on the ground in the daytime. Many brightly colored butterflies are found congregating around puddles. Although some moths may be flushed from their resting places and netted by day, and some are diurnal, the most productive collecting method is to catch or trap them when they are attracted to light. Bait collecting also is a standard technique for some groups (see p. 16). Most lepidopterous larvae feed on live plants, some species feeding on or in the leaves and others boring into the stems, seeds, or other parts of the plant. The larvae of many species are very host-specific, starving rather than feeding on a plant other than the preferred species. A few larvae are scavengers on dead plant or animal matter, including woolen clothing.

Lepidoptera have long been especially popular for rearing, and the breeding and hybridization of butterfly species have become highly developed. No special equipment or skills are necessary to rear most foliage-feeding species from eggs. Many adult female moths will lay eggs freely if confined in a small plastic box, glass jar, or other container with a piece of the food plant or some crumpled paper. For large moths, a paper bag or shoebox can be used. Keep a piece of damp paper towel or sponge in the container to maintain the humidity, especially if the specimens are reared in an air-conditioned or heated building. A water-soaked raisin will provide nourishment for the female as well as moisture for those species feeding as adults.

Most butterflies require large, brightly illuminated cages containing an ample supply of the natural food to induce them to ovipost. Females caught outdoors will nearly always be fertile, and their eggs will hatch in a week or 10 days unless the species is one that overwinters in the egg stage. If the food plant is unknown, a trial-and-error procedure must be used by offering the newly hatched larvae bits of leaves from a variety of plants that are thought to be possible hosts.

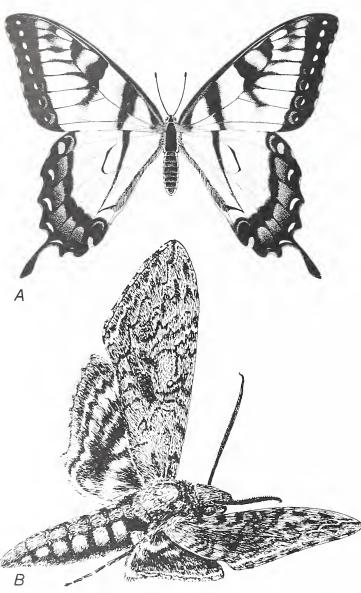


Figure 52. A, Butterfly, Papilio glaucus L., order Lepidoptera, family Papilionidae; B, tomato hornworm moth, Manduca quinquemaculata (Haworth), order Lepidoptera, family Sphingidae.

Rearing cages made of screening are appropriate for large moths and most butterflies, but caterpillars require little air, and most species may be reared successfully in closed plastic or metal containers. Excessive moisture need not be a problem if the containers are kept at a reasonably constant temperature and the bottom is lined with a clean paper towel or blotting paper. Only dry foliage should be used as food; caterpillars need only the water they get by eating fresh leaves, and tight containers keep the leaves fresh for several days. Cleanliness is essential, and uneaten leaves and the accumulation of frass at the bottom must be removed at regular, often daily, intervals. When mature, the larvae should be provided with suitable conditions for pupation. For example, species that pupate in the ground may need several inches of slightly dampened peat, sand, or sawdust. Species that pupate in leaf litter often will do well if allowed to crawl into the folds of a crumpled paper towel.

Preserving the various larval stages is often desirable so that they may be identified by positive association with adults reared from the same brood. Larvae should be killed by immersing them for a minute or so in nearly boiling water. They may then be preserved in alcohol. Large specimens should have the alcohol changed after the first 24 hours because the body fluids will have diluted it. As an alternative to alcoholic preservation, larvae may be freeze-dried or inflated and then mounted on pins.

Perhaps the most difficult problem in collecting adult Lepidoptera is to avoid rubbing off the wing scales. For large specimens, a widemouthed killing jar is recommended, with the killing agent of sufficient strength to kill or stun the insect immediately. Do not place large beetles or other large insects in the same jar with butterflies and moths, and do not crowd too many Lepidoptera into one jar. Some collectors prefer to remove the specimens soon after they have been killed and place each specimen temporarily in a separate, labeled envelope; this procedure is especially useful for butterflies. Such papered specimens later can be relaxed and spread. For permanent collections, adult Lepidoptera should be pinned with the wings spread in the standard manner (see p. 32) so that all important features of wing pattern and structure may be seen. Never glue adults to points or place them in alcohol. Pin small microlepidoptera with minutens, spread, then double mount. Papered specimens or specimens left too long in a killing jar must be placed in a relaxing chamber before the wings can be spread, and even fresh specimens are easier to handle if placed overnight in a relaxing chamber (see p. 24).

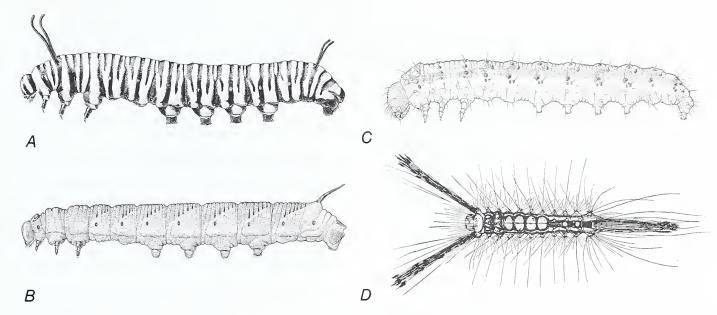


Figure 53. Larvae (caterpillars) of *A*, monarch butterfly, *Danaus plexippus* (L.), order Lepidoptera, family Danaidae; *B*, tomato hornworm, *Manduca quinquemaculata* (Haworth), order Lepidoptera, family Sphingidae; *C*, corn ear-

worm, *Heliothis* zea (Boddie), order Lepidoptera, family Noctuidae; *D*, whitemarked tussock moth, *Orgyia leucostigma* (J. E. Smith), order Lepidoptera, family Lymantriidae.

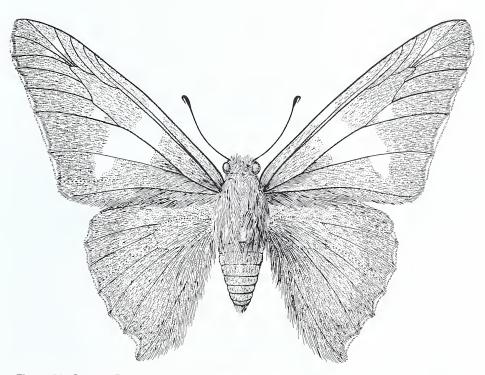


Figure 54. Skipper, *Epargyreus* sp., order Lepidoptera, family Hesperiidae.

Diptera ("True" Flies, Mosquitoes). Adult Diptera (fig. 55) are soft-bodied insects with only the front pair of wings; the hindwings are modified into a pair of slender, knobbed structures called halteres. Other orders may have species with only one pair of wings, but no other insects have knobbed halteres. (The halteres or hamulohalteres of winged male scale insects are not knobbed and usually are tipped with one or more hooked setae. Most winged male scale insects also differ from flies in having a single long, stylelike process at the end of the abdomen.) The mouthparts of Diptera, whether in the form of a tube, a pair of spongy lobes, or a sharp beak, are fitted for sucking liquids. Metamorphosis is complete. The larvae (fig. 56) are commonly called maggots. They lack true legs, but some species have one or more pairs of prolegs. In some primitive families, the larvae have a distinct head capsule, and the pupae are free living. In more advanced flies, as in the muscoid families, no head is apparent on the larvae, which often pupate inside the last larval skin, known as a puparium.

The order Diptera contains approximately 120 families and in North America alone over 18,000 described species. Some are beneficial, such as those that pollinate flowers or prey on or parasitize other insects; other species are serious crop pests or important vectors of diseases. The greatest number of flies, however, probably are neither beneficial nor harmful to man, but often are collected and studied because of the unusual ecological niches they occupy.

Habitats of Diptera are numerous and varied. Perhaps the most unusual are the seeps of crude petroleum in which larvae of some ephydrid larvae (shore flies) (fig. 56, *B*) develop or the hot springs in which some stratiomyid larvae are found. A rain-filled can by the side of the road may harbor mosquito larvae (family Culicidae) (fig. 56, *C*), and stones in a fast-flowing stream may hold simuliid (black fly) larvae. A dipper or small aquatic net is useful in collecting such larvae. A sweep net or aerial net can be used effectively to collect the adults, which generally remain near the water.

Although most dipterous larvae are aquatic or semi-aquatic, some species live in plant tissues, mining leaves, forming galls, or feeding in the stems or roots. By placing the host plant, or infested parts of the host plant, in a rearing container, it may be possible to keep the larvae until they pupate and the adult flies emerge. Similarly, adult tachinids and other parasitic flies can be obtained by keeping the parasitized host insect in a rearing container. Because some species normally pupate in the soil, place an inch or so of moist sand or moss in the bottom of the cage.

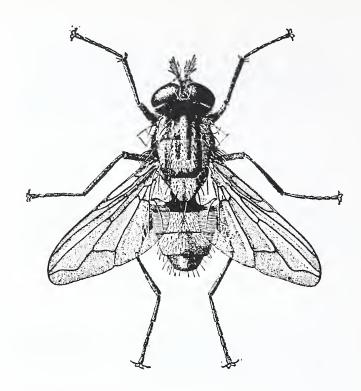


Figure 55. Striped horsefly, *Tabanus lineola* (Fab.), order Diptera, family Tabanidae.

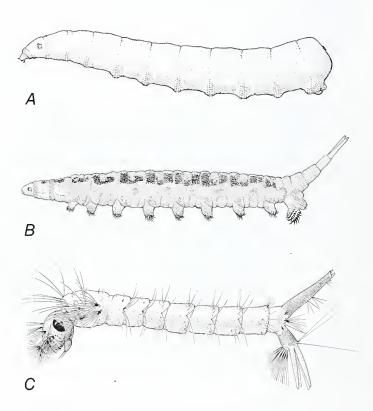


Figure 56. Larvae of *A*, house fly, *Musca domestica* L., order Diptera, family Muscidae; *B*, shore fly, *Hydropyrus hians* (Say), order Diptera, family Ephydridae; *C*, northern house mosquito, *Culex pipiens* L., order Diptera, family Culicidae.

Flies that are scavengers on decaying animal and vegetable matter may be collected in bait traps. Chemical baits that attract some specific groups of flies are available commercially. Light traps or flight traps, such as the Malaise trap (see p. 11), used with or without a bait, usually are extremely successful in capturing adult flies.

Some flies are ectoparasites of bats, birds, and other animals. Many of these parasitic flies are wingless and can be collected readily from the host with forceps or an aspirator.

Most flies should be killed dry and mounted on pins, minutens, or card points, if possible within a few hours after they have been killed. Remember that newly emerged adults may be teneral and should be held alive until the wings and colors fully develop. Small flies, if not mounted on minutens or points very soon after collecting, should be placed in alcohol and run through the Cellosolve-xylene series (see p. 25). No flies should be kept dry-layered in boxes or envelopes; their heads, antennae, and legs are far too prone to become detached. Dipterous larvae should be killed in nearly boiling water and preserved in alcohol. For permanent collections, mosquito larvae are preferably mounted on microscope slides. Some adult flies also can be preserved satisfactorily in alcohol, but most should never be placed in alcohol. If adults of many families are immersed in liquid, the scales, hairs, or bristles critical for identification may come loose and the specimens will become useless.

Siphonaptera (Fleas). Adult fleas (fig. 57) are small, wingless insects with the body greatly compressed laterally, usually heavily sclerotized, and armed with combs of spines or bristles. The antennae are short and inconspicuous; the legs are long, spiny, and equipped for jumping. Adult fleas are parasitic on warm-blooded vertebrates and have mouthparts of the piercing-sucking type with long, well-developed palpi. Metamorphosis is complete. Flea larvae are legless, with long bristles over most of the body. The larvae do not suck blood but feed on organic debris in and about the nest or habitation of the host. In North America, there are about eight families in the order; current authorities disagree as to the exact number. Fleas such as the cat flea, Ctenocephalides felis (Bouché), and the dog flea, C. canis (Curtis), are well known as household pests, but fleas are of primary importance to humans because of the potential of some species to transmit bubonic or sylvatic plague, endemic typhus, and other serious diseases.

Fleas most commonly infest animals that live in nests or burrows and seldom are found on cattle, deer, or other hoofed animals. Rodents and other small mammals are almost always heavily infested with fleas, which can be collected by shooting or trapping the host and immediately picking or brushing the fleas from the fur. Because fleas are active, it may be necessary to place the host in a jar or bag with a few drops of liquid killing agent to stun or kill the fleas before examining the fur. Placing the host animal in a refrigerator for an hour or longer also will slow the activity of the fleas. If immediate examination of the host is not possible, it should be placed in a bag (each host animal in a separate bag) to confine the fleas, which tend to leave a dead host.

Birds also have fleas, but the fleas that attack birds usually are collected most readily by examining the nest soon after it has been abandoned in the summer. In every instance, the species of bird that made the nest should be noted on the data label. The nest can be teased apart or placed in a Berlese funnel or sifter to collect the adult fleas and immature stages. Because the latter cannot be identified with certainty without associated adults, some of the larvae or pupae should be placed in jars with a quantity of the nest material and reared. For successful rearing, it is essential to maintain the humidity as high as would normally be found in a nest. The burrows or nests of mammals also should yield numerous adult and immature fleas. Adults and larvae may be killed and preserved satisfactorily in alcohol, but for permanent collections, it is preferable to mount adult fleas on slides. Be sure to include the name of the host on the slide label.

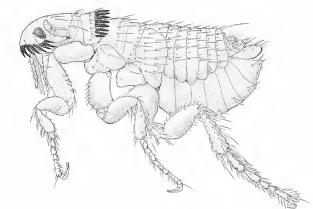


Figure 57. Cat flea, Ctenocephalides felis (Bouché), order Siphonaptera, family Pulicidae.

Hymenoptera (Sawflies, Ants, Wasps, Bees). Hymenoptera may be wingless (fig. 58, A) or winged (fig. 58, B), with two pairs of membranous wings. The forewings are larger, and the hindwings are usually coupled to them with a series of little hooks fitting into a fold in the forewing. Mouthparts are equipped for chewing or for chewing and sucking and may be modified into a tonguelike structure. Metamorphosis is complete. Most hymenopterous larvae have a distinct head and are legless and maggotlike (fig. 59, A), but the larvae of sawflies (fig. 59, B) have thoracic legs and prolegs. The latter lack crochets, a character that may be used to separate these larvae from the larvae of Lepidoptera. The order Hymenoptera consists of 2 suborders and 71 families.

The habits of Hymenoptera are diverse, ranging from free-living, plant-feeding forms such as sawflies, to parasitic forms such as ichneumonids and braconids, to social forms such as some wasps, ants, and bees. Many bees are important pollinators of plants, and some are valued for the honey they produce. The parasitic forms are of ever-increasing importance as biological control agents. The plant-feeding forms include some of the most destructive defoliators of forest trees.

Because the habits are so diverse, collecting methods vary. Sawfly adults (fig. 60), so-called because of their sawlike ovipositor, may be caught in interception traps such as the Malaise trap or swept from low vegetation. Most sawfly larvae are external feeders on vegetation, but some species mine the leaves or leaf petioles, and a few species cause galls. Outbreaks of sawflies may occur occasionally in a given area and offer collectors an excellent opportunity to obtain samples large enough to allow some specimens to be preserved as immatures and others to be held in rearing cages until adults emerge. Procedures used in rearing Lepidoptera (see p. 74) can be used for rearing sawflies. Sawfly larvae may be preserved in alcohol, inflated, or freezedried. Many sawflies attach their cocoons to leaves or twigs of the host plant, but larvae of some species drop to the ground and pupate in the soil or leaf litter. For this reason, 5-8 cm of moist soil or peat moss should be kept on the bottom of the rearing cages. The adults that emerge may be killed in alcohol but later should be removed and pinned, preferably with the wings extended.

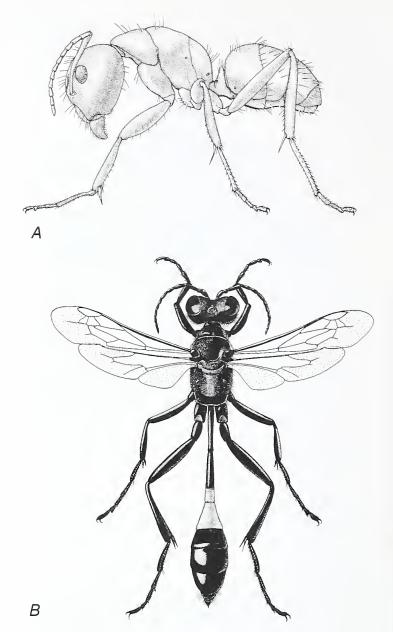


Figure 58. *A,* Common ant, *Camponotus castaneus* (Latreille), order Hymenoptera, family Formicidae; *B,* thread-waisted wasp, *Ammophila urnaria* Dahlbom, order Hymenoptera, family Sphecidae.

Parasitic wasps will often be found in Malaise traps. in Berlese samples, and in samples taken in a sweep net: however, for accurate association of the parasites with their hosts, it is essential that they be reared. [It is the policy of the Biosystematics and Beneficial Insects Institute not to accept parasitic Hymenoptera for identification unless the host insect has been identified at least to genus.] Rearing is also one means of associating the sexes with certainty, since males and females in many species differ greatly in appearance. It is important to include on the collection label not only the scientific name of the host insect, but also the life stage of the host from which the parasite emerged. If the specimens are determined to be polyembryonic (more than one individual produced from one egg), this also should be noted on the label. If possible, the remains of the host should be retained and mounted below the parasite on the pin. Parasitic wasps too small for direct pinning should be glued to points. Do not mount microhymenoptera on minutens.

In mounting parasitic Hymenoptera, the antennae, legs, and wings should be arranged so as not to obscure characters on the body needed for identification. Be careful with ichneumonids, however, that the long wings and antennae do not point upward and thus possibly may be broken when the pin is handled. Tiny specimens, those under 2 mm, may be cleared and mounted on slides or kept in alcohol. Scientists in the Systematic Entomology Laboratory prefer that specimens of the Ichneumonoidea be killed and preserved in 95 percent alcohol; other parasitic Hymenoptera may be preserved satisfactorily in 70-80 percent alcohol.

Gall wasps and the galls from which they emerge should be preserved together, since identification of many species is based on the gall. If galls are placed in a pinned collection, make sure that they are pinned securely, with additional brace pins on each side. Use care when labeling specimens associated with a particular gall. Not all insects that emerge are gall producers; some may be parasites of the gall wasps or inquilines.

Ant nests also may yield other kinds of insects besides ants; many of them mimic ants. In collecting ants from a nest, use an aspirator or Berlese funnel and try to obtain specimens of each caste. It may be necessary to return to a nest periodically through the year to find the males or winged females. Ants may be collected and preserved in alcohol, but for a permanent collection, larger specimens should be pinned, and specimens under 5 mm should be glued to points.

Aculeate or stinging wasps and bees may be caught in Malaise traps or by sweeping. See page 4 on how to remove stinging insects from a net. If specimens are taken from a nest, try to collect the nest also. If this is not practical, make a note or sketch of the approximate measurements of the nest. Do not overlook the insects that the wasps may have stored in cells in the nest.

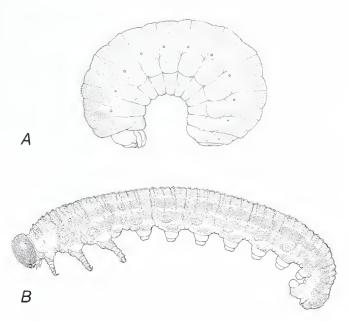


Figure 59. Larvae of *A*, megachilid bee, order Hymenoptera, family Megachilidae; *B*, European pine sawfly, *Neodiprion sertifer* (Geoffroy), order Hymenoptera, family Diprionidae.

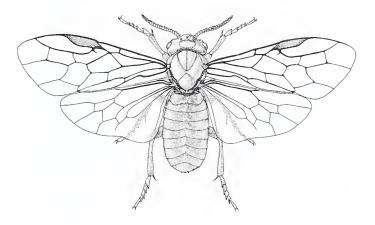


Figure 60. Adult pine sawfly, *Neodiprion sertifer* (Geoffroy), order Hymenoptera, family Diprionidae.

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Appendix

Formulas

Distilled water should be used in these formulas if available, but rainwater or bottled drinking water is satisfactory. "Parts" is by volume.

AGA (Alcohol-Glycerin-Acetic Acid) Solution

,	4,10
Commercial ethanol (ethyl alcohol)	- 5 - 1

Barber's Fluid

49
19
7
1

Hoyer's Medium

Chloral hydrate	20
Water	5
Gum arabic (granules)	3
Glycerin	2

Dissolve gum arabic in water at room temperature. Add chloral hydrate and allow to stand for a day or two until all solids have dissolved. Add glycerin. Filter through glass wool. Store in glass-stoppered bottle.

Essig's Aphid Fluid

Lactic acid	20
Glacial acetic acid	4
Phenol (saturated H ₂ O solution)	2
Distilled H ₂ O	1
-	

KAAD (Kerosene-Acetic Acid-Dioxane) Solution

Commercial ethanol (ethyl alcohol)	10
Glacial acetic acid	2
Kerosene	1
Dioxane	1

Mix in the order given. For very soft-bodied larvae, use half as much kerosene or less. Dioxane may be omitted.

Sample Mounting Procedures

Parts

The following procedures for mounting certain insects and mites for scientific study are preferred by the Systematic Entomology Laboratory (ARS, USDA). A few of the chemicals indicated by an asterisk (*) in these procedures are hazardous. Carefully investigate their properties to insure their safe use.

Mounting Aphids, Scale Insects, and Aleyrodids. Specimens of aphids, scale insects, and aleyrodids cannot be pinned because of their small size and their tendency to shrivel. The following procedures are recommended:

- (1) Place specimen in 10 percent potassium hydroxide (KOH)* solution and heat gently until body contents are softened, or leave in KOH solution at room temperature for up to 48 hours.
- (2) Remove specimen from KOH and place in 70 percent ethanol for 5 minutes. Note for aleyrodids: If black specimens have not turned brown at this point, bleach them in peroxide-ammonia solution (1 drop ammonia to 6 drops hydrogen peroxide) until brown. Next place in 95 percent ethanol for 5 minutes; then proceed to step 7.
- (3) Remove from 70 percent ethanol and place in Essig's Aphid Fluid. Make incision halfway across body between second and third pairs of legs. Then squeeze a few times to remove and flush out body contents. If feasible, one or two well-formed embryos should be left in bodies of aphids. Excess wax may be removed by placing specimens in tetrahydrofuran*. (NOTE: This is a hazardous chemical and should be used under exhaust hood and in a very well-ventilated place to avoid inhalation of fumes.)
- (4) Remove from Essig's fluid and place in 95 percent ethanol for 5 minutes.
- (5) Remove from the ethanol and place in acid fuchsin stain for about 5 minutes or until properly stained, then place in 70 percent ethanol for 5 or more minutes to remove excess stain.
- (6) Remove from 70 percent ethanol and place in 95 percent ethanol for 5 minutes.
- (7) Remove from 95 percent ethanol and place in clove oil; leave for 5 minutes or until specimen appears nonshiny, dull, and flat.
- (8) Remove from clove oil and place specimen dorsum upward on a slide in a drop or two of Canada balsam. If mounting three or more specimens on one slide, place them in a row right to left with one specimen ventral side up. Keep all specimens neatly horizontal with heads pointed in same direction.
- (9) Put cover slip in place and attach labels, preferably so that they may be read with heads of specimens toward you.

Mounting Thrips. For a detailed study, mount thrips in Canada balsam as described here. Place each specimen by itself centrally on a slide with wings, legs, and antennae spread for easy observation of structures. Most specimens should be cleared for optimal appearance of surface detail, but a few should be left in their natural color by omitting steps 2 and 3. Rapid identifications may be made from temporary mounts in glycerin or Hoyer's medium, but they usually cause distortion. Excess or used fluids may be removed at each step with a pipet.

- (1) Soak specimen for 24 hours in clean 60 percent ethanol to remove collecting fluid.
- (2) Macerate in cold 5 percent sodium hydroxide[⋆] solution for 30 minutes or up to 4 hours for especially dark specimens.
- (3) Wash briefly in 50 percent ethanol and then leave in 60 percent ethanol for 24 hours.
- (4) Dehydrate through a series of ethanol solutions: 70 percent for 1 hour, 80 percent for 2 hours, and 100 percent for 10 minutes (change alcohol once). Place in clove oil until clear (30 seconds to 10 minutes). Spread appendages carefully at each stage. Dehydration and clearing may be promoted by puncturing thoracic and abdominal membranes in one or two places with a fine needle.
- (5) Place ventral side uppermost on 13-mm cover slip in Canada balsam, then lower slide onto cover slip. This method is easier to control than the usual method of lowering cover slip onto slide with forceps.
- (6) Use two labels on slide, one at each side of specimen, with host, locality, altitude, date, and collector's name on right-hand label and determination and sex data on left-hand label.
- (7) Cure in oven at 40° C within a few minutes of preparation, or leave for up to 6 weeks for complete curing.

Mounting Mites Other Than Eriophyids. Mites are most easily mounted if collected in AGA solution (see p. 94). Mount those collected in 70-80 percent ethanol on slides as soon as possible. The following procedures do not apply to mites of the family Eriophyidae:

- (1) Place drop of Hoyer's medium (see p. 94) in center of clean 1- by 3-inch microscope slide.
- (2) Remove mite directly from host, or pour specimens from collecting vial into small casserole, watch glass, or petri dish. Avoid pouring too much fluid from vial into dish; the less fluid, the easier it is to pick out the mites. The mites may be removed from the host or from the fluid by dipping a needle into the Hoyer's medium on the slide and then quickly touching the mite with it.

- (3) Place single specimen in medium on slide. Press specimen to surface of slide and spread all legs laterally. Most mites should be mounted dorsoventrally, but males of many species, such as those of the Acaridae and Tetranychidae, should be mounted laterally to allow examination in profile of the specifically characteristic aedeagus. Some mites may require a small body puncture to eliminate the contents. Heavily pigmented mites may be cleared in a solution of lactophenol before mounting. The solution may be heated to hasten clearing.
- (4) With forceps, carefully place clean, small (13-mm or smaller) cover slip on mite in Hoyer's medium. Gently press cover slip with forceps to hold mite in position.
- (5) Place slide on hotplate set at 65° C and remove it rapidly when single bubble forms in Hoyer's medium. Avoid more bubbling or mite will be displaced.
- (6) Turn slide so anterior part of mite is directed toward you.
- (7) Place label on slide to right of cover slip; host, locality, collector, date, and serial number data should be shown on this label.
- (8) Place slide in oven at 45°-50° C for 24 hours or longer to cure and solidify the Hoyer's medium. The slide must be kept horizontal until the medium is firm and there is no danger of the cover slip moving.
- (9) After removing slide from oven, ringing cover slip with additional Hoyer's medium helps to prevent the mount from drying out.

BBII LOT NO. BBII PRIORITY U.S. DEPARTMENT OF AGRICULTURE AGRICULTURAL RESEARCH SERVICE BIOSYSTEMATICS AND BENEFICIAL INSECTS INSTITUTE DATE Sender's Reference No. **IDENTIFICATION REQUEST** DATE IDENTIFICATION REQUIRED (month, day, year) If less than two months, explain below NOTE: • Please type or print all information. TOTAL NUMBER SENT Do not write in shaded areas. Give explanations where requested in "Remarks" section at bottom of form. Vials: Slides Attach additional pages if (and only if) more space is needed. Other: NAME & COMPLETE MAILING ADDRESS OF SENDER (Include Zip code) RETURN TO (If other than sender) (Include Zip code) SOURCE **REASON FOR IDENTIFICATION** (Check and complete as appropriate) su 🗆 State University ☐ ARS Biological control OS Other State 1 Scientific name of target pest. APHIS-PPQ PU \Box Private University AQ 2 General quarantine or biocontrol research \Box FS ES IN Individual 3 ldentity of host of natural enemy DD 🔲 U.S. Military co Commercial Organization 4 Recovery of released natural enemy OF ☐ Other Federal FN ☐ Non-U.S. 5 Suspected contaminant in culture ☐ CICP CL ☐ State Agricultural Agency 6 Quarantine reference collection LEVEL/TYPE OF IDENTIFICATION NEEDED 7 D Voucher specimen of field release ☐ Family ☐ Genus □ Species 8 Holding living material pending identification ☐ Damaging crop, plants — identify host plants: Positive or negative verification of ecological group: ☐ phytophagous parasitic □ predaceous c Suspected pest of regulatory concern—give details below other: ☐ saprophagous aphidophagous Stored product pest — commodity affected: OTHER INFORMATION REQUESTED — Will be supplied as conditions allow Livestock, wildlife, or domestic animal pest - host: as determined by taxonomist. ☐ Danger to human health a Pest status d Best reference ☐ Household pest — damage: **b** Geographical distribution d ldentifying characteristics Possible immigrant—new to: ☐ Reference collection — for: f Other (Specify) ☐ Usual hosts (prey) ☐ Survey—explain in detail below SOURCE OF PROJECT SUPPORT Regional ☐ Thesis problem — describe project below ☐ M.S. ☐ Ph.D ☐ ARS ☐ APHIS ☐ FS ☐ CSRS project no. ☐ Other — explain below ☐ Hatch ☐ EPA ☐ NIH □ NSF ☐ FAO ☐ USAID ☐ Other (Specify) SPECIMEN DISPOSITION VOUCHER SPECIMEN(S) INCLUDED TELEPHONE REPORT REQUESTED If yes, give number - include area code and extension How marked? Return ☐ Place in National Collection Requests are handled at the discretion of BBII Keep part Will be ☐ Other (Specify) of series deposited in DESCRIPTION OF PROJECT - Include Project Title and name of Project Leader. (Reference previous communications pertaining to this submittal) REMARKS (Explanations, tentative identification, etc.) FOR BBII USE DATE RECEIVED NO. LABEL SORTED PREPARED DATE ACCEPTED CC's PART I-BBII COPY ARS-748 (5/86)



Agricultural Research Service

Beltsville Area

Biosystematics and **Beneficial Insects** Institute Beltsville, Maryland 20705

IDENTIFICATION REQUEST-INFORMATION/INSTRUCTIONS

The Biosystematics and Beneficial Insects Institute (BBII) is the U.S. Federal agency responsible for providing identifications and other taxonomic services for insects and related organisms. We are pleased to be of assistance; however, it is appropriate that you determine if sources of identifications are available in your area or country before submitting material.

1. Use this form for submission of each lot—not for individual specimens. Retain Part III for your records. Send Part I in advance and Part II enclosed with your submittal (or send Parts I and II with your submittal) to:

> Dr. Lloyd Knutson, Director Biosystematics and Beneficial Insects Institute Building 003, Room 1, Beltsville Agricultural Research Center-West Agricultural Research Service, USDA Beltsville, Maryland 20705 U.S.A.

- 2. Each specimen, whether pinned, in a vial, or on a slide, must be labeled as to—specific locality (country, state, or other political subdivision, and city or pertinent local landmark), date of collection, name of collector, family, genus, and species names of host (if known), and voucher number (if appropriate). Each specimen should have a unique number to facilitate reporting of identifications. See reverse side for preparation of specimens. Please see our statement on parasitic Hymenoptera for information on our requirements on host data for parasitic wasps.
- 3. A brief description of your project will enable us to place the proper priority on your request. Early submittals, and submittals of small lots as studies progress, will ensure faster service.
- 4. Lots relating to agricultural interests are given highest priority. Requests for information on hosts, distribution, identification characteristics, literature references, etc., are generally answered at the option of the taxonomists based on their evaluation of the request and time available. To provide the requested information the taxonomist often must spend considerable time and effort. Please request only the information that is critically needed.
- 5. Inquiries on the status of an identification request may be made by— Correspondence to Lloyd Knutson at the above address Telephone - Mary Lacey (301) 344-3041 or (301) 344-4451 Telemail - MLACEY

Technical inquiries on identifications or responses made by the taxonomists should be sent directly to them.

- 6. Specimen(s) that represent new species, new host records, or new distribution records may be retained at the discretion of the taxonomist for placement in the U.S. National Collection of Insects in the Smithsonian Institution. Approximately 25,000 specimens are added to the National Collection every year as a result of the BBII identification service. The opportunity to retain specimens of interest is one way in which our collaborators can be recompensed for their assistance.
- 7. Users of this service should appropriately cite the taxonomist in their publications and reports. If the identifier cannot be given after the name of the taxon (e.g., in tables or lists), a footnote or other means of acknowledgment should be used. Proper formatting, as appropriate, is as follows:

Name of taxonomist, Systematic Entomology Laboratory, Agricultural Research Service,

U.S. Department of Agriculture

Name of taxonomist, Department of Entomology, Smithsonian Institution Cooperating entomologists at other institutions should be listed in similar format.

8. We would greatly appreciate your sending reprints of publications and other documents in which our identifications are used to Lloyd Knutson at the above address and to the taxonomist(s) who provided the determination(s).

Form ARS-748A (5/86)

The insect and mite taxonomic service program of the Biosystematics and Beneficial Insects Institute (BBII), with the aid of entomologists in the Smithsonian Institution and other cooperators, provides identifications of about one-third million specimens per year (about 30,000 scientific names, or 17,000 consignments or lots) for an extremely wide variety of users. Unfortunately, some specimens must be returned unidentified because the poor condition of a specimen may prevent its identification, the classification of a group may need or be undergoing revision, the quantity of material may be too large for us to accept, or the request may not be in line with our priorities. We also lack specialists or cooperators for some groups (see list below).

Immature stages are accepted for identification, but the classification of immature forms is so poorly known in many groups that even identification to family is not possible. For this reason we urge all collectors to rear some of the specimens and submit adults with the associated immature stages.

Preparation of Specimens Submitted for Identification . . . Because of a lack of sufficient technical assistance to prepare specimens for identification, we are including the following instructions to help you prepare specimens in a condition suitable for study.

PINNED—Most specimens should be pinned; those too small or fragile for direct pinning should be double mounted on minuten nadeln or carefully glued to paper points. Glue the point to the *right side* of the specimen, using care that the glue does not conceal critical characters. Do not glue tiny *moths* or *flies* to points; use minuten nadeln. Specimens should be pinned while fresh. *Moths* should be submitted with wings spread. Examination of properly prepared genitalia is necessary to identify many insects to genus or species level. Specific instructions for preparing genitalia will be supplied upon request. Puparia, pupal skins, cocoons, etc., should be placed in a gelatin capsule or glued to a card either separately or pinned below the adult.

SLIDE MOUNTS—Submit mites, fleas, thrips, aphids, whiteflies, psychodid flies, most scales, and mosquito larvae on microscope slides, if you can prepare good slides. This will enable us to identify the material more quickly. Some trichogrammatid and mymarid wasps and other minute insects must also be mounted on slides. Larval ticks are acceptable as slide mounts if they are not engorged. Specific instructions for slide-mounting the above groups will be supplied upon request.

ALCOHOL—Submit the following specimens in alcohol: *ichneumonid wasps, mayflies, bat flies,* all *soft-bodied insects* (including all *larvae* and *pupae*), and most *insects under 2 mm* in length, except as indicated below. Also, specimens in the groups listed under *Slide Mounts* should be preserved in 70% alcohol if not slide-mounted. Adult *whiteflies* may be submitted in alcohol, but identification of this stage usually is not possible. **Never** submit adults of the following in alcohol: *moths, bees, true bugs, psychodid, or all other flies except minute Nematocera flies.* **Place Only One Kind of Insect in Each Vial.** Use neoprene-, rubber-, or silicone-stoppered vials rather than screw-capped or shell vials. Use clear glass vials of sufficient size to allow the use of forceps or an eye dropper to remove the specimens; however, if the vial is too large, very small specimens will be difficult to find. Do not use methanol or formalin solutions; use 70, 80, or 95% ethanol if possible. Isopropanol, the drugstore variety, is adequate only for temporary storage. To prevent dilution of the alcohol and subsequent decomposition of specimens, fresh alcohol must be placed in the vials within 24 hours after *initial immersion* of specimens. All vials containing soft-bodied insects should be shipped exclusive of all air bubbles. Insert a paperclip or pin with the stopper to eliminate all air bubbles; then remove the paperclip or pin.

PREPARATION of SPECIFIC FORMS—Kill *larv*ae by placing them in boiling water or in an alcohol-glacial acetic acid mixture, then transfer them to 70% alcohol. *lchneumonid* wasps and *mayflies* should be killed and preserved in 95% ethanol; *thrips* should be killed and preserved in AGA (9 parts 70% alcohol:1 part glycerine:1 part glacial acetic acid). Nymphal, adult, and engorged larval *ticks* should be preserved in 70-80% ethanol.

DRY, UNMOUNTED—If whiteflies and diaspidid scales are not mounted on slides, they should be submitted on host plants placed between pieces of dry paper towel, blotters, or other absorbent paper. Do not place specimens belonging to these families in plastic bags.

PILLBOXES — Pillboxes and matchboxes are NOT acceptable containers for submitting insects, but they may be used to submit associated plant samples, galls, or similar material. Soft tissue paper or cellucotton, not cotton, should be used in such boxes.

Identification Capability... Adults and immatures of all groups of insects and related arthropods are accepted for identification by BBII taxonomists, except those listed below. Groups marked by an asterisk are accepted but referred to cooperators outside the Institute. Identification of these is at the discretion of the cooperator. Groups not marked by an asterisk are usually returned unidentified. With appropriate justification, we may attempt to identify some of these groups or may provide names and addresses of other experts.

*Ephemeroptera Protura *Anoplura *Neuroptera Thysanura *Collembola *Mallophaga *Trichoptera *Odonata *Psocoptera Diplura *Mecoptera *Plecoptera *Siphonaptera **INSECT GROUPS** COLFORTERA Pedilidae *Dryomyzidae Empididae HEMIPTERA Hesperioidea Amphizoidae Anthicidae *Incurvarioidea *Micropterygoidea ingidoidea Psephenidae Rhysodidae Ephydridae Buprestidae Carabidae 'Heleomyzidae 'Lauxaniidae *Nepticuloidea *Papilionoidea Silphidae HOMOPTERA Sphaeriidae Coccoidea Cicindelidae Slaphylinidae Mydidae Psyllidae *Sesiidae Sphingidae Nemestrinidae *Ptychopteridae Cleridae COLLEMBOLA Dryopidae LEPIDOPTERA (adults) Tineioidea Dytiscidae DIPTERA Scenopinidae Tortricoidea Bombycoidea *Tanyderidae *Tipulidae Elmidae Acroceridae rponomeutoidea Contomorphoidea Georyssidae Apioceridae *Cossoidea Zygaenoidea Trichoceridae Asilidae Blephariceridae Eriocranioidea Haliplidae ·Trixoscelididae *Henjalnidea Hydraenidae Bombyliidae Hydrophilidae Camillidae 1 There are also several minor families for which no specialist is available Hydroscaphidae Celyphidae USDA material will be accepted.
 ARS Biological Control material will be accepted. Lagriidae *Ceralopogonidae Limnichidae Chironomid *Coelopidae Melandryidae 4 True butterflies (Papilionoidea) - from outside North America only will be identified it (1) voucher specimens Mordellidae Nitidulidae Noteridae Oedemeridae may be retained, (2) the specimens have been reared or parasitized, and (3) all specimens are fully labeled, in-'Culicidae *Curtonotidae cluding food plant data Diastatidae NON-INSECT GROUPS *Drosophilidae Diplopoda (millipedes) Pedipalpida ('whip scorpions") 'Araneida (spiders) *Argasidae ("softbacked ticks" *Phalangida ("daddy longlegs") Gastropoda (snails & slugs) Chelonethida (pseudoscorpions) Scorpionida (scorpions) Hydrachnellae (water mites) Chilopoda (centipedes)
Crustacea (includes sowbugs & pillbugs) 'Ixodidae ("hardbacked ticks") Oligochaeta (earthworms) Solpugida (solpugids) Symphyla ("symphylans")

No permit can be issued to move live plant pests or noxious weeds until a cation is received (7 CFR 330 (live plant pests) or 7 CFR 360 (noxious w U.S. DEPARTMENT OF AGRICULTURE ANIMAL AND PLANT HEALTH INSPECTION SERVICE PLANT PROTECTION AND QUARANTINE BIOLOGICAL ASSESSMENT SUPPORT STAFF HYATTSVILLE, MARYLAND 20782									
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STANDARD SAFEGUARDS OF PERMIT

- All pests must be shipped in sturdy, escape-proof containers.
- Upon receipt of pests, all packing material and shipping containers shall be sterilized or destroyed immediately after removing pests.
- 3. Pests shall be kept only within the laboratory or designated area at the permittee's address.
- No living pests kept under this permit shall be removed from confined area except by prior approval from State and Federal regulatory officials.
- Without prior notice and during reasonable hours, authorized PPQ and State regulatory officials shall be allowed to inspect the conditions under which the pests are kept.
- 6. All pests kept under this permit shall be destroyed at the completion of the intended use, and not later than the expiration date, unless an extension is granted by this issuing office.
- 7. All necessary precautions must be taken to prevent escape of pests. In the event of an escape, notify this office.

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